Spoilage and biopreservation of veal meat

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INTRODUCTION

Microbial ecosystem of veal meat has not been well described especially spoilage flora despite this product could be highly perishable. In 2011, the ANR ECOBIOPRO(1) project allowed to identify the microbial ecosystem of fresh and spoiled ground veal meat using pyrosequencing of the 16S rDNA gene. *Brochothrix thermosphacta/campestris*, *Leuconostoc gasicomitatum*, *Lactococcus piscium*, *Lactobacillus sakei* and *Leuconostoc citreum* were selected as the main potential spoilage species. Challenge-tests were conducted with these species in order to study their spoilage potential. The impact of bioprotective cultures was evaluated on ionized veal meat artificially contaminated with spoilage species and then on naturally contaminated veal meat.

APPROACH

1. **The spoilage potential was studied for each species separately. Cocktail of 1 to 4 strains were inoculated at initial concentration level of 3 log cfu/g in veal meat previously decontaminated by ionisation (12 kGy). Artificially contaminated veal meat was stored under modified atmosphere during 12 days at 8°C. Se nsory evaluations were performed during the storage.**
2. **The main spoiling species *Ln. gasicomitatum, Ln. citreum* and *Lc. piscium* were therefore selected to study the impact of 4 bioprotective cultures (BC) in veal meat previously decontaminated by ionisation (12 kGy). Each bioprotective culture was inoculated at initial level of 6 to 7 log cfu/g with and without spoilage species (SS). Artificially contaminated veal meat was stored under modified atmosphere during 4 days at 4°C and 8 days at 8°C. Sensory eval uations were performed during the storage.**
3. **The impact of 2 biopreservative cultures was then studied on the global ecosystem (by pyrosequencing analysis) in naturally contaminated veal meat. Biopreserved veal meat was stored under modified atmosphere during 4 days at 4°C and 8 days at 8°C. Sen sory evaluations were performed during the storage.**

1. IMPACT OF BIOPROTECTIVE CULTURES ON SPOILAGE SPECIES

Lc. piscium, *Ln. gasicomitatum* and *Ln. citreum* which were associated to vinegar odor and/or discoloration of the meat were chosen to study the impact of bioprotective cultures on their growth and spoilage activities in ionized veal meat.

Bioprotective cultures

- *Lactobacillus curvatus* **(LC)**
- *Staphylococcus carnosus* + *Pediococcus acidilactici* **(SC-PA)**
- *Lactobacillus sakei* + *Staphylococcus xylosus* **(LS-SX)**
- *Lactobacillus sakei* **(LS)**

- **Spoilage of ionized veal meat was observed after 8 days of storage (D8) for SC-PA, LC and LS and after 12 days (D12) for LS-SX when inoculated without spoilage species (Table 1).**
- **Ionized veal meat inoculated with spoilage species (without bioprotective cultures) were spoiled at D8 for *Lc. piscium* (discoloration) and at D12 for *Ln. citreum* and *Ln. gasicomitatum* (Table 1).**
- **The use of LS strain allowed to reduce spoilage due to *Ln. citreum* (odor and color; Table 2).**
- **With LC starter, odor defects were less important for *Ln. citreum* and *Ln. gasicomitatum*.**
- **No change was observed for *Lc. piscium* with the 4 bioprotective cultures studied (Table 2).**

SPOILAGE DATE	+ LS	+ SC-PA	+ LC	+ LS-SX	without bioprotective cultures
Lactococcus piscium	D12	D8	D8	D8	D8
Leuconostoc citreum	D8	D8	D8	D8	D12
Leuconostoc gasicomitatum	D8	D12	D8	D8	D12
without spoilage strains	D8	D8	D8	D12	Blank Not spoiled at D12

Table 1 : Date of spoilage of ionized veal meat artificially contaminated with spoilage species (SS), bioprotective cultures (BC) or with SS + BC and stored at 8°C

COLOR	+ LS	+ SC-PA	+ LC	+ LS-SX
L. piscium				
L. citreum				
L. gasicomitatum				

ODOR	+ LS	+ SC-PA	+ LC	+ LS-SX
L. piscium				
L. citreum				
L. gasicomitatum				

Table 2 : Impact of bioprotective culture (BC) on spoilage (odor and color defects) of ionized veal meat artificially contaminated with spoilage species (SS)
(green: less important spoilage with BC ; orange: little improvement of quality of meat with BC; red: equal spoilage with and without BC)

2. IMPACT OF BIOPROTECTIVE CULTURES ON THE NATURAL ECOSYSTEM AND SPOILAGE OF VEAL MEAT

LS strain (*L. sakei*) and LC starter (*L. curvatus*) were chosen to study the impact of bioprotective cultures on spoilage of three batches of naturally contaminated veal meat stored under modified atmosphere during 4 days at 4°C and 8 days at 8°C.

- **Veal meat inoculated with LC starter were more spoiled than not inoculated meat after 11 days of storage due to discoloration (Fig.1)**
- **Veal meat inoculated with LS strain were less spoiled than not inoculated meat after 11 days of storage *(p value 0.007)* (Fig.1)**
- **No change in odor was observed between blank and assays.**

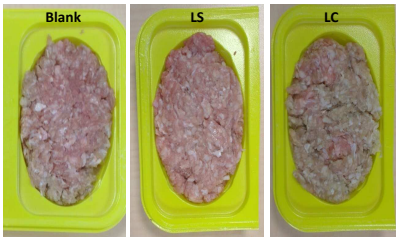

Figure 1: Aspect of artificially contaminated veal meat without bioprotective culture (Blank) and with LS (*L. sakei*) strain or LC (*L. curvatus*) starter after storage during 4 days at 4°C and 8 days at 8°C under modified atmosphere

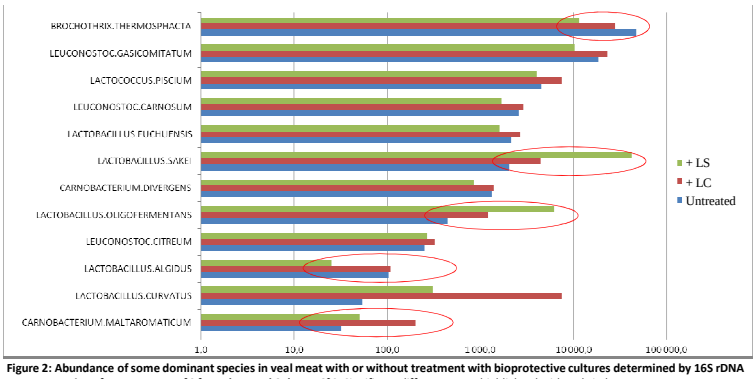

Figure 2: Abundance of some dominant species in veal meat with or without treatment with bioprotective cultures determined by 16S rDNA pyrosequencing after storage at 4°C for 4 days and 8 days a 8°C. Significant differences are highlighted with red circles.

The microbial ecosystem of veal meat, untreated and treated with *L. sakei* (LS) or *L. curvatus* (LC), has been investigated by sequencing 16S rDNA after a 11 days storage (4 days at 4°C + 7 days at 8°C) and compared (Fig.2).

- **Contrary to LC starter, LS strain abundantly colonized the meat.**
- **The beneficial effect of LS is not due to its presence itself but rather to the changes which it induced in the abundances of other species: spoiling species *B. thermosphacta* and *Lb. algidus* are significantly inhibited whereas *Lb. oligofermentans* growth is stimulated.**
- **The ecosystem of veal meat treated with LC is similar to that of the untreated control. Even so, *C. maltaromaticum* is more abundant in presence of LC.**

CONCLUSION

This study allowed to confirm the spoilage potential of dominant species isolated from veal meat at the end of the shelf-life. Sensory evaluations showed that *Lc. piscium*, *Ln. gasicomitatum* and *Ln. citreum* were associated to vinegar odor and discoloration of the meat. The impact of four bioprotective cultures on their growth and spoilage activities was tested on ionized veal meat. *Lactobacillus sakei* (LS strain) and *Lactobacillus curvatus* (LC starter) reduced the spoilage after storage at 8°C. In na turally contaminated veal meat, only LS strain allowed a better preservation after 11 days of storage at 4°C then 8°C. The results confirmed that bi opreservative cultures could improve the shelf-life of fresh veal meat stored under modified atmosphere at cold temperature.

Analysis of the veal meat ecosystem after treatment with bioprotective cultures demonstrated that even inoculated at high levels, starters did not substantially alter the flora of the product. However, some significant changes in the abundance of a few dominant species could explain the beneficial effect of the bioprotective strain LS on the spoilage of veal meat.

Changes in the lactic acid flora and some other species were also shown to significantly impact the ability of the bioprotective cultures to colonize the meat (data not shown). An extensive product characterization is therefore necessary for the implementation of an effective biopreservation.

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