# HALOTOLERANT BACTERIA INVOLVED IN DRY-CURED HAM SURFACE DEFECTS

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## Background

Surface defects of microbiological origin have been affecting dry-cured ham production for long time and are still far from being completely wiped out. Recent statistics indicate that 40% of these defects are located near the point of exit of the femoral vein. This alteration is easily identifiable by olfactive analysis, but microbiological analysis often fails to demonstrate the cause.

## Objectives

The aim of this study was to determine the microbial agents responsible for the "vein defect" in spoiled dry-cured ham, by comparing the typical microflora of the normal hams to the microbial populations isolated from the spoiled hams.

## Methods

64 samples were analysed: 34 from spoiled and 30 from normal hams. Hams were obtained from different meat processing plants. Slices approximately 0.2 cm in thickness were aseptically removed from the area near the point of exit of the femoral vein (2 cm wide).
The following microbiological analyses were performed: aerobic mesophilic count in tryptone soya agar (TSA, Oxoid; 30°C for 72 hours); enterobacteria in violet red bile glucose agar (VRBGA, Oxoid; 30°C for 48 hours); *Micrococcaceae* in mannitol salt agar (MSA, Oxoid; 30°C for 72 hours); yeasts in malt extract agar (MEA, Oxoid; 30°C for 72 hours) and halotolerant bacteria count in tryptone soya agar containing 6% NaCl (w/w) (TSSA; 30°C for 72 hours).
Halotolerant strains grown on MSA and on TSSA were isolated and identified. The strains were characterised using following tests: Gram stain, cell morphology, oxidase and catalase activity. Biochemical characterization and final identification of isolated strains were carried out by the Biolog System (EIAFOSS) following producer's instructions.

## Results and discussion

The average values of microbial counts, expressed as $\log_{10}$ cfu/g, are shown in Table 1 and 2 for spoiled and unspoiled hams, respectively.

Table 1 – Main values ($\log_{10}$) of microbial counts in spoiled hams

| | Aerobic mesophilic count | *Micrococcaceae* | Yeasts |
|---|---|---|---|
| Average | 7.83 | 6.99 | 4.89 |
| Standard deviation | 0.71 | 1.39 | 1.06 |
| Min. | 5.56 | 4.18 | 3.08 |
| Max | 8.86 | 8.48 | 6.96 |

Table 2 – Main values ($\log_{10}$) of microbial counts in unspoiled hams

| | Aerobic mesophilic count | *Micrococcaceae* | Yeasts |
|---|---|---|---|
| Average | 7.38 | 6.54 | 5.27 |
| Standard deviation | 1.04 | 1.87 | 1.11 |
| Min. | 4.78 | 2.11 | 2.85 |
| Max | 8.68 | 8.86 | 6.63 |

No significative differences between aerobic mesophilic, *Micrococcaceae* and yeasts counts in spoiled or in unspoiled hams were found. All these microbial groups together represent the microflora naturally present on the muscle surface of hams in the maturing phase.
Only 20 % of spoiled hams and 10 % of the unspoiled hams were positive for *Enterobacteriaceae*, and the contamination levels were always below $10^4$cfu/g (Figure 1).
Other halotolerant bacteria than *Micrococcaceae* were found in the samples taken from both spoiled and unspoiled hams; the research was therefore focused on the isolation and identification of these microrganisms.
Bacteria belonging to the specie *Brochothrix thermosphacta* were isolated from 20% of spoiled samples after 4 months maturing, at contamination levels of about $10^3$ cfu/g; these bacteria were no more detected in spoiled hams after 8 months maturing and at the end of maturing phase. *B. thermosphacta* was isolated, at level of about $10^3$ cfu/g, from 46.1% of unspoiled hams before 8 months maturing (Figure 2). Halotolerant bacteria different from *B. thermosphacta* were present in 50 % of the spoiled hams at levels ranging from $10^5$ to $10^9$ cfu/g (Figure 3). Since these bacteria were almost absent in unspoiled hams, it was assumed they could have been responsible for the onset of the defect.

Figure 1 – Distribution of positive samples for *Enterobacteriaceae*

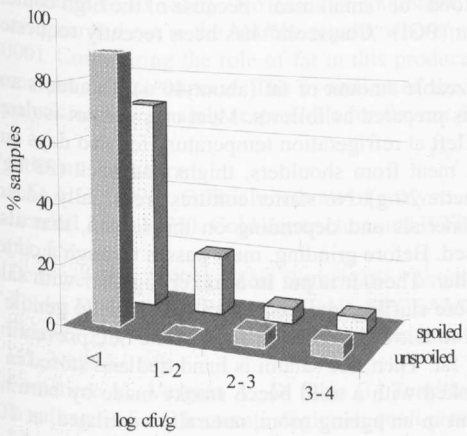



Figure 2 – Distribution of positive samples for *B. thermosphacta*

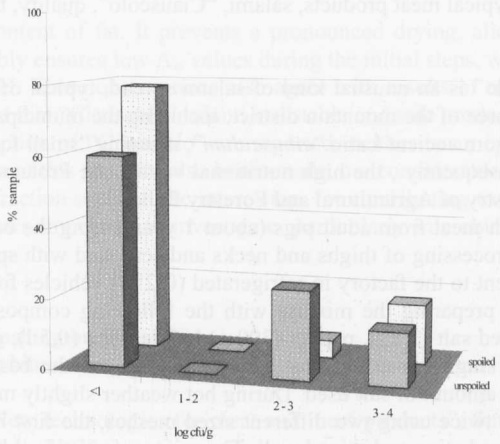
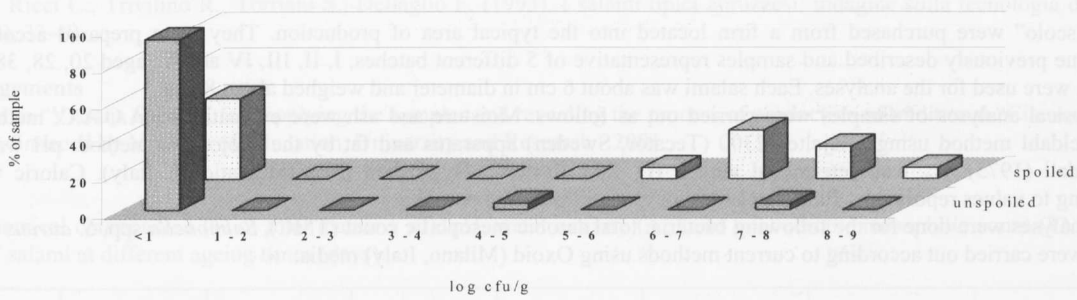



Figure 3 – Distribution of positive samples for halotolerant bacteria




Among the bacteria grown on MSA and TSSA 26 strains were isolated and identified: 16 were Gram negative and 10 Gram positive. All Gram negative bacteria proved positive to the oxidase test and belonged to the genus *Vibrio*. Six of the Gram positive strains proved negative to the catalase test and were identified as *Carnobacterium*. The other four strains, catalase positive, belonged to *Corynebacterium* genus.

**Pertinent literature**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