Food Development & Production

Impact of nitrite on the microbiological quality of a Portuguese *Salpicão*

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Introduction

The development of healthier traditional product formulations is required by new trends in the food industry[1,2]. The curing of meats is disseminated throughout the world, being firmly established in traditional gastronomies. Meat curing is widespread throughout the world and is an integral part of traditional gastronomy. The use of nitrite salts is paramount in suppressing lipid oxidation and reduce growth of pathogenic bacteria. This activity is especially significant when inhibition of *Clostridium botulinum*[2] is necessary, inhibiting vegetative cell proliferation and germination of spores[3,4,5]. While this is widely recognised, the impact of nitrite on the total microbiota of fermented meats is not yet fully understood. The ingestion of excessive nitrite has been correlated with an increased risk of cardiovascular pathologies, diabetes, blood diseases, and cancers[2,6]. As a result, a popular adverse reaction to the use of nitrite salts has been trending[6].

With NGS (Next-Generation Sequencing) techniques, detecting changes in microbial communities can be performed with unparallel specificity. Therefore, this work aimed to analyse the development of microbiological characteristics, as a result of nitrite elimination, throughout the various processing phases of *Salpicão*.

Methods

Samples of each phase of production (raw meat T0; before stuffing T1; middle of curing process T2; finished product T3; middle of shelf-life T4; and end of shelf-life T5) were collected and total DNA was extracted following the Qiagen DNeasy® mericon® food kit protocol. This extraction was performed independently in two different lots. Amplification of the 16s rRNA gene, specifically the hypervariable V3/V4 region, was performed. PCR products were sequenced by the Illumina MiSeq® sequencer.

Results and Discussion

- Significant variation between lots was observed.
- Bacteria of the *Lactobacillus* genus dominated the microbiota of the later phases in both samples.
- The reduction observed in *Listeriaceae* presence might be a result of the production of bacteriocins by *Lactobacillus* species.
- Lactic Acid Bacteria cultures as starters could hinder pathogenic proliferation.
- *Pseudomonas*, *Lactobacillus*, and *Weissella* were not affected in the presence of nitrite.
- *Brochothrix*, and *Salmonella* appeared susceptible to nitrite activity.
- *Clostridium* was found in lower abundance in samples with nitrite (0.02%) than in nitrite-free samples (0.23%).
- Nitrite seems to be effective in preventing *Clostridium* spp. growth

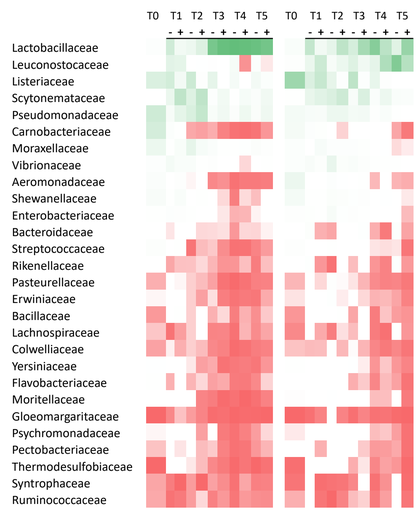

Figure 1 – Heatmap of the total microbiota, at the Family level. Legend: Green – High density; Red – Low density; White – Percentile 50; (+) - with nitrite; (-) – without nitrite

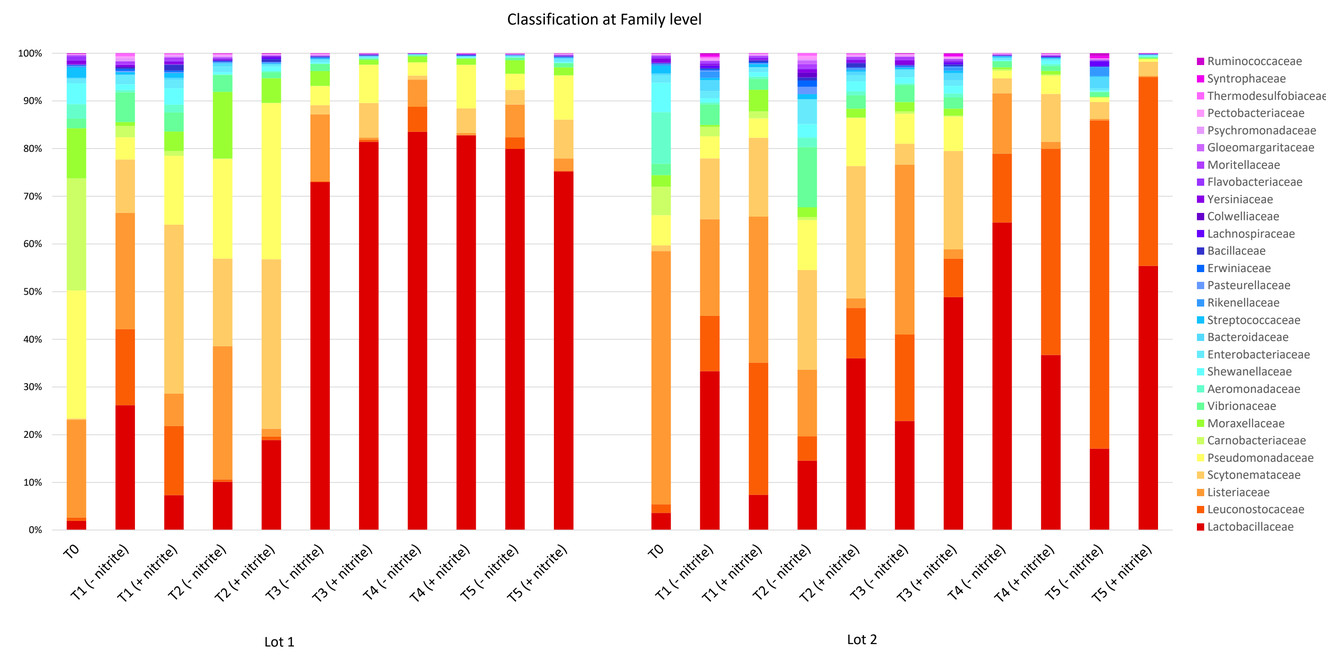

Figure 2 – Variation of the total microbiota throughout processing, with and without nitrite, at the Genus level.

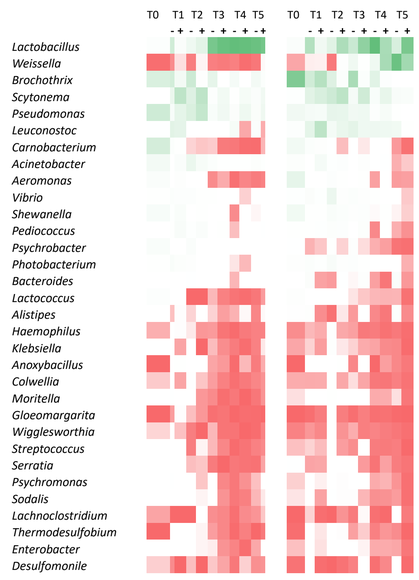

Figure 3 – Heatmap of the total microbiota, at the Genus level. Legend: Green – High density; Red – Low density; White – Percentile 50; (+) - with nitrite; (-) – without nitrite

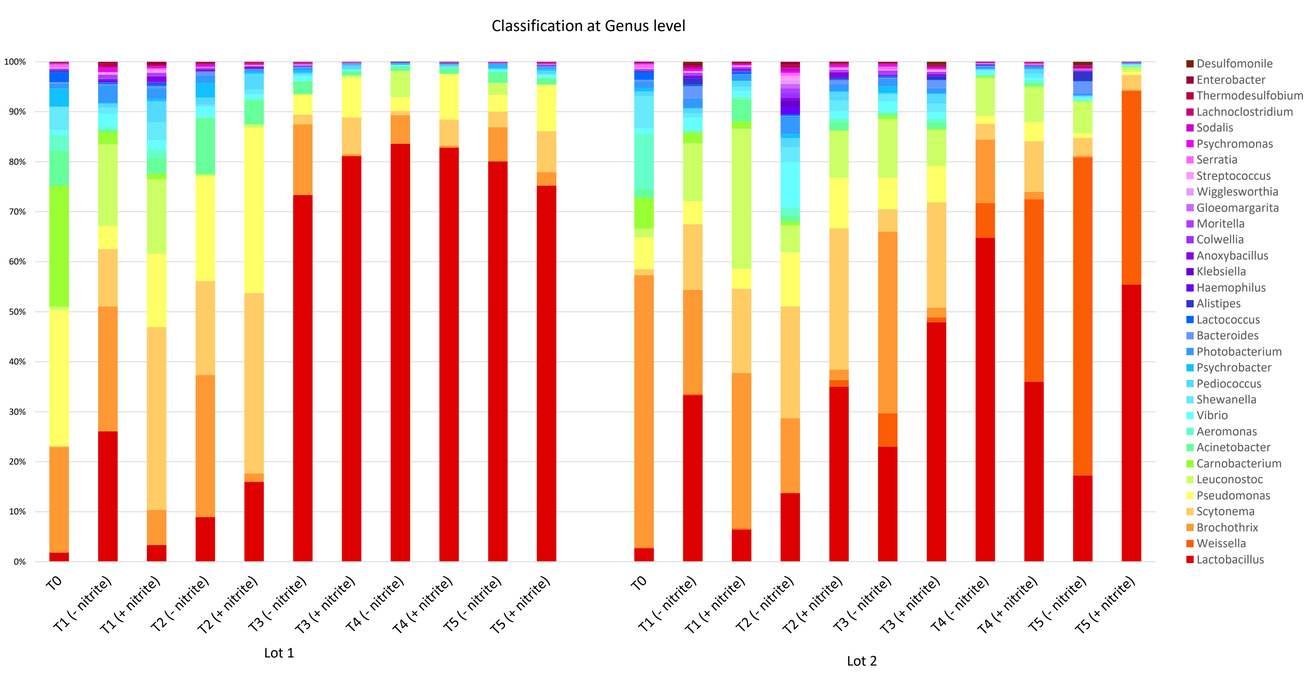

Figure 4 – Variation of the total microbiota throughout processing, with and without nitrite, at the Genus level.

Conclusions

- There is considerable variability between different lots of *Salpicão*.
- Nitrites have an unquestionable impact on the microbial composition of the product.
- Sensorial and stability alterations might occur due to the impact of nitrites on the microbiota.
- Lactic Acid Bacteria dominate the microbiota of the later processing stages.
- *Listeria* decreases throughout processing, possibly due to LAB bacteriocins.
- Products without nitrites have heightened levels of *Clostridium.*

Organized by:

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