Luminous Photobacterium spp. in chicken meat spoilage: growth patterns and genotypic diversity

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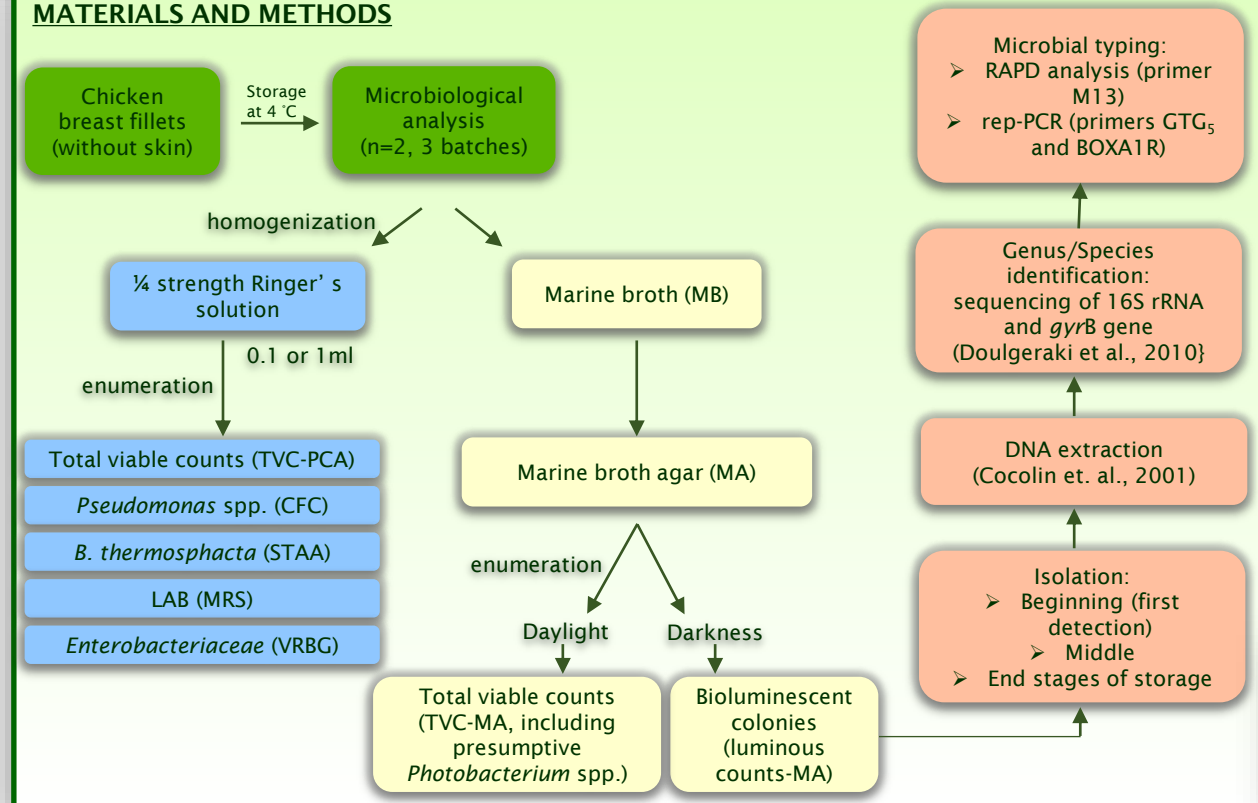
INTRODUCTION

Microbial chicken meat spoilage has been traditionally attributed to genera *Pseudomonas*, *Brochothrix (B.) thermosphacta*, *Enterobacteriaceae* and lactic acid bacteria (LAB). Yet, recent metagenomic analyses on spoiled chicken meat revealed the presence of genus *Photobacterium* at significantly high abundances.

OBJECTIVE

This study was conducted to a) assess the presence and growth patterns of luminous *Photobacterium* spp. on refrigerated chicken breast fillets and b) explore and describe their genotypic biodiversity.

MATERIALS AND METHODS



RESULTS AND DISCUSSION

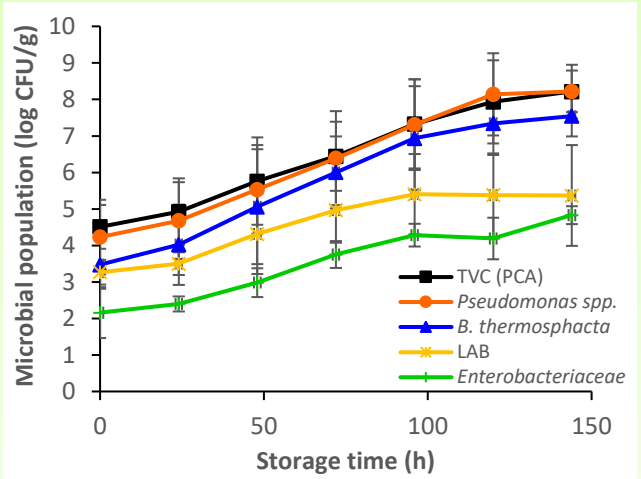


Figure 1. Microbial counts of TVC (PCA), *Pseudomonas* spp., *B. thermosphacta*, *Enterobacteriaceae* and LAB on chicken breast fillets stored at 4 °C (mean ± standard deviation, n=6).

- The microbial association of chicken breast fillets consisted mainly of *Pseudomonas* spp., B. *thermosphacta*, LAB and *Enterobacteriaceae*.
- The population of all microbial groups increased during storage at 4 ˚C, with *Pseudomonas* spp. being the dominant microbiota followed closely by *B. thermosphacta*, while LAB and *Enterobacteriaceae* reach lower maximum bacterial populations.

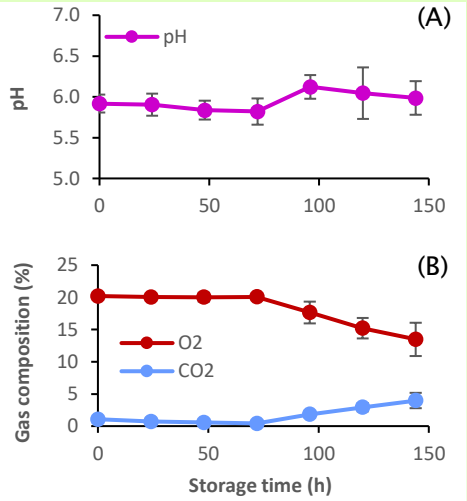


Figure 2. Changes on pH values (A) and O_2, CO_2 concentrations (B) on chicken breast fillets and packaging headspace during storage at 4 °C for each of the three experimental batches (mean ± standard deviation, n=6).

- pH remained remained relatively unchanged.
- A decline on the O_2 concentration along with an increase on the CO_2 concentration after 72 h (due to the non-permeable to gasses plastic packaging film and the increase on CO_2 concentration due to the microbial metabolic activity.

Acknowledgment

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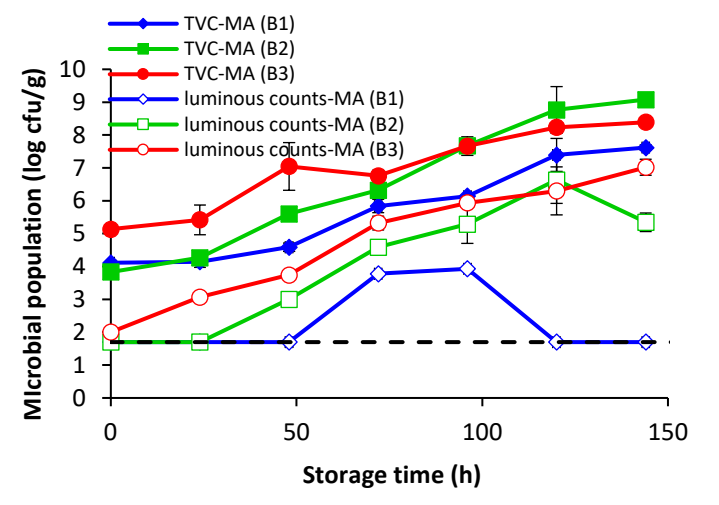

Figure 3. Evolution of TVC (bold symbols) and luminous bacteria (open symbols) on chicken breast fillets stored at 4°C for each of the three experimental batches (B1-B3), as enumerated on MA medium (mean ± standard deviation, n=2). Dashed line indicates the detection limit of the enumeration method (DL=1.7 log CFU/g).

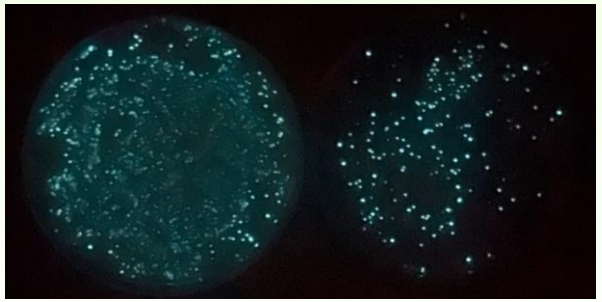

Figure 4. Luminous bacteria on chicken breast fillets stored for 6 days at 4 °C as enumerated on MA.

- TVC populations enumerated on MA were higher than luminous counts and similar to those enumerated on PCA, and ranged from 7.6 ± 0.1 to 9.1 ± 0.0 log CFU/g (mean 8.2 ± 0.6 log CFU/g) depending on the batch (Figure 3). The difference between luminous and total counts on MA suggest that the population of bioluminescent bacteria may have been underestimated due to the overgrowth of the TVC-MA (Figure 4).
- Bioluminescent bacteria were scarce at the beginning of storage and were detected in only one out of three batches at a population of *ca.* 2.0 log CFU/g.
- As storage time proceeded, they were encountered at higher populations ranging from 5.4 ± 0.3 log CFU/g to 7.0 ± 0.2 log CFU/g for the for the second (B2) and third (B3) batch, respectively. In the case of first batch (B1), luminous bacteria were sporadically encountered throughout storage at populations of up to 3.9 log CFU/g.

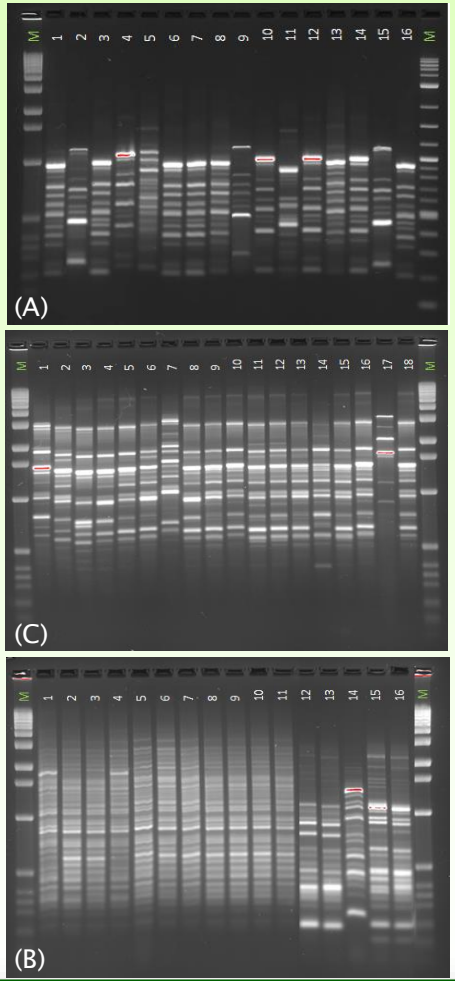

Figure 5. Representative RAPD profiles generated with primer M13 (A), and REP-PCR profiles generated with primers GTG_5 (B) and BOXA1R (C). M, 1 Kb DNA molecular marker.

- A total of 91 presumptive *Photobacterium* spp. isolates were isolated from chicken breast fillets during storage (Figure 4).
- DNA fingerprinting reveals so far diverse DNA fingerprints of bioluminescent bacterial isolates (Figure 5). Similarities on RAPD and PER-PCR fingerprint patterns will be analyzed with Bionumerics software in order to discriminate strains.
- 16S rDNA gene sequencing of selected isolates confirms that they belong to *Photobacterium* genus. Phylogenetic analysis of combined 16SrRNA and *gyr*B gene sequences will further distinguish isolates within the *Photobacterium* genus.

CONCLUSION

Results of the present study highlight the potent role of *Photobacterium* spp. to chicken meat spoilage and reveal the significant genetic biodiversity of *Photobacterium* isolates.

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