

P-02-03**Combination of essential oils and phenolic acids against *salmonella* for food safety of sliced cooked ham (#276)**

Jessica A. Feijó Corrêa, João V. Santos, Alberto G. Evangelista, Renata E. F. Macedo, Fernando B. Luciano

Pontifícia Universidade Católica do Paraná (PUCPR), Graduate Program in Animal Science, Curitiba, Brazil

Introduction

Salmonella is one of the most reported agents associated to enteric diseases, presenting elevated risk and economical losses in the food industry. Natural antimicrobials have gained relevance through the years, and the combined application of essential oils and phenolic acids has shown potential results to control food pathogens [1,2]. This study aimed to assess the effects of several combinations of phenolic acids (PAs) and essential oil components (EOCs) on the growth of *Salmonella*, and, thereafter, evaluate the best combination to control *Salmonella* in sliced cooked ham.

Methods

A four strains pool of *Salmonella* Enteritidis (CRIFS 1016, 56301, 33SUSP and 9SUSP) was cultivated in Müeller Hinton broth (pH 6). The minimum inhibitory concentrations (MICs) of the PAs trans-ferulic, 4-hydroxybenzoic, o-coumaric and syringic, and of the EOCs allyl isothiocyanate, cinnamaldehyde and carvacrol were determined. Then, the fractional inhibitory concentrations (FICs) of the substances in the 12 possible combinations (PA+EOC) were evaluated combining decreasing fractions of each MIC. The method was microdilution as described by CLSI [3]. The most effective combination was elected for application in ham.

For that, 8 x 7 x 0.2 cm cooked ham slices were spread-inoculated with 60 µL of a 10⁸ colony-forming units (CFU)/mL suspension of the pool and air dried in safety cabinet. For the treatment, 8 x 7 cm filter papers were previously impregnated with the PA at concentrations equivalent to 5 and 10x the FICs (5X and 10X). Papers were positioned on top of the inoculated surface of slices, which were packaged in modified atmosphere packaging bags. Another filter paper was used for the EOC application, also at 5X and 10X, and positioned next to the slice. Bags were heat sealed and incubated for 7 d, 4 °C. Positive and negative controls were also prepared – inoculated/non-treated and non-inoculated/non-treated, respectively. Color (parameters L*, a*, b*, C* and h) and pH were measured at the storage times 0, 1, 4 and 7 d. Likewise, microbiological analyses were conducted as described by Meira et al. [2]. Experiments were performed twice in triplicate. Data were analyzed by one-way ANOVA followed by Tukey's test (P≤0.05) in GraphPad Prism 5.04.

Results

Determined MICs and FICs are shown in Tables 1 and 2, respectively. PAs and EOCs showed enhanced activity when applied together. Cinnamaldehyde and o-coumaric acid combination was elected for evaluation in sliced ham due to its lower FIC values and similar chemical characteristics. Slices

treated with the elected combination did not show color nor pH alterations with statistical significance compared to control (data not shown). Also, microbiological results (Figure 1) imply the tested concentrations were not effective on reducing *Salmonella* population through the storage period.

A significant reduction in *Salmonella* population was observed on day 1 by 10X treatment, however this reduction was not maintained during the analysis. As observed in previous studies, PAs and EOCs combination show great potential against foodborne pathogens [1,2].

Conclusion

Different application methods must be developed to achieve an effective and continuous delivery in the matrix, aiming to establish bacterial inactivation during the shelf life. Since color and pH are not altered, there is possibility of commercial use without affecting consumer acceptability.

Acknowledgments

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References

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Notes

FICs (EOC + PA, mM)				
+	trans-Ferulic	4-Hydroxybenzoic	o-Coumaric	Syringic
AITC	1.26 + 5.15	1.26 + 7.24	1.26 + 6.09	1.26 + 10.1
Cinnamaldehyde	3.78 + 5.15	1.89 + 3.62	1.89 + 3.05	1.89 + 5.05
Carvacrol	6.66 + 5.15	3.33 + 3.62	6.66 + 6.09	6.66 + 10.1

Table 2. FICs determined for PAs and EOCs combinations against a 4-strain pool of Salmonella.

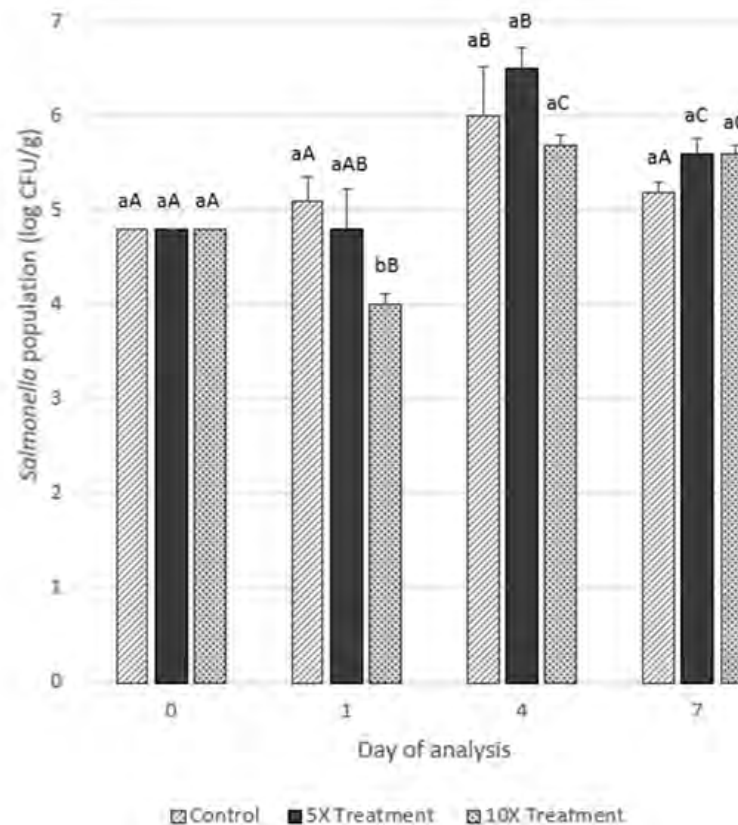


Figure 1. Residual population of Salmonella spp. in ham treated with cinnamaldehyde and o-coumaric. Different lowercase and uppercase letters indicate statistical difference between days and between treatments, respectively ($P \leq 0.05$).

Notes

Essential oils components and phenolic acids	MIC (mM)
Allyl isothiocyanate	1.26
Cinnamaldehyde	3.78
Carvacrol	6.66
trans-Ferulic acid	5.15
4-Hydroxybenzoic acid	7.24
o-Coumaric acid	6.09
Syringic acid	10.1

Table 1.
MIC of PAs and EOCs against a 4-strain pool of Salmonella.

Notes