

Escherichia coli inactivation in apple puree treated by high-pressure processing guaranteeing safe food product

Enrique Pino Hernández¹, Marco Alves¹, Carlos A. Pinto², Marta B. Evangelista^{1,3}, Jorge A. Saraiva²

¹INOV.LINEA - Agri-food Technology Transfer and Valorization Center, TAGUSVALLEY – Science and Technology Park, 2200-062, Abrantes, Portugal

²LAQV-REQUIMTE, Chemistry Department, University of Aveiro, Campus Universitário de Santiago, 3810-193, Aveiro, Portugal

³LEAF—Linking Landscape, Environment, Agriculture and Food Research Center, Associated Laboratory TERRA, Instituto Superior de Agronomia, Universidade de Lisboa, Tapada da Ajuda, 1349-017 Lisboa, Portugal

Introduction

High-pressure processing (HPP) is used for the nonthermal (5°C - 20°C) pasteurization of foods and beverages (Pino-Hernández *et al.* 2022). This technology has demonstrated the potential to be used in minimally processed foods for extending their shelf-life and producing safe products for the food industry (Pinto *et al.* 2020). Apples are the main fruit in terms of production in the European Union (EU). Data reported by FAOSTAT (2022) show that in the EU, apple production has been increasing raising from 9,638 t (2017) to 11,833 t (2020). Purée is one of the most important apple product in the market (Landel *et al.* 2010). This product can be utilized in food for babies or the elderly, having a higher market value due to the practicality and comfort when compared to apple fresh. This work investigated the inactivation of *Escherichia coli* (ATCC 25922) (*E. coli*) in *Reineta* apple puree treated by HPP and evaluated the effects of this technology on total aerobic mesophilic, moulds and yeasts and inoculated *E. coli* loads for 30 days under refrigerated storage (5 °C) at 70 % relative humidity (RH).

Materials and Methods



•Physicochemical analyses:

Before and immediately after HPP, physicochemical characteristics such as colour, °Brix, and pH were evaluated. Value $\Delta E^* \geq 5$ indicates the limit beyond which the human eye can perceive strong color differences.

•Microbial analyses:

Apple puree was inoculated with ~ 7 log CFU/g of *E. coli* and the bacteria load and was evaluated by plating serial dilutions in MCA Agar. Native microorganisms (total aerobic mesophilic bacteria, moulds and yeasts) were determined by plating serial dilutions in PCA and RBC agar, respectively. These analyses were assessed at 0, 10, 20, and 30 days of storage.

Results and Discussions

The results showed that immediately after HPP at 400 and 500 MPa, *E. coli* was inactivated at least by 5.0 log cycles in apple puree from *Reineta* variety (Figure 1). These treatments could be considered mild pasteurization in accordance with the FDA's 5-log reduction criteria (FDA, 2004).

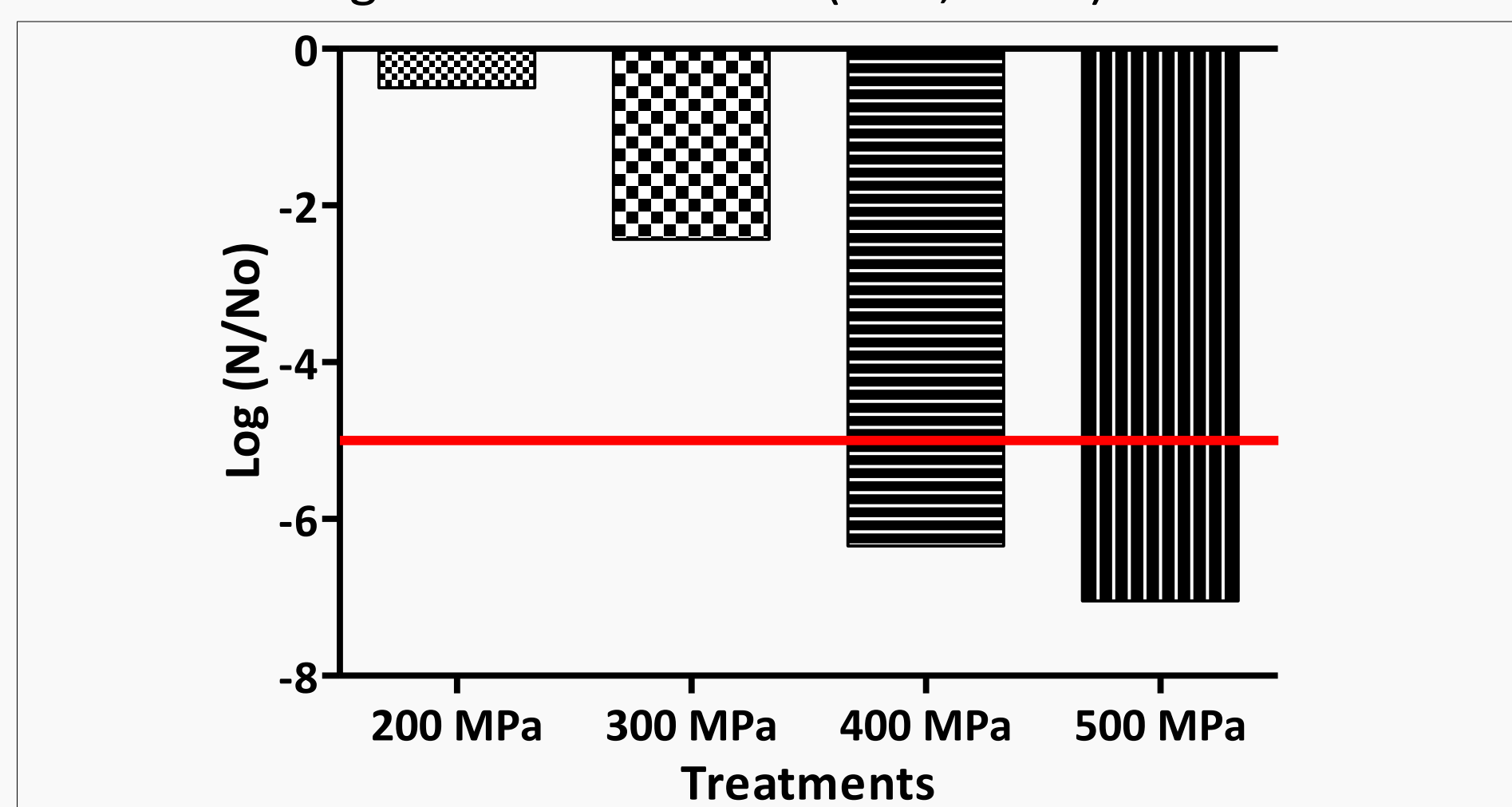


Figure 1. Reduction of *Escherichia coli* in HPP-treated *Reineta* apple puree. N denotes the microbial load measured for each treatment and N_0 the initial microbial load (7.0 log CFU/g).

Table 1, show that no negative changes on physicochemical characteristics of the product were found after HPP. ΔE^* color differences were not strongly perceptible to the human eye.

Initial microbial loads in apple puree were effectively reduced by pressures ≥ 300 MPa to non-detectable levels. Nonetheless, at 300 MPa after 30 days of storage, the total aerobic mesophilic bacteria, moulds and yeasts increased.

Table 1. Changes in the physicochemical parameters and total mesophilic bacteria, moulds and yeast populations in control and HPP samples puree.

Treatments	Storage time (days)	TPC (log CFU/g)	M&Y (log CFU/g)	ΔE^* Color	°Brix	pH
Control	0	3.5	5.5	--	13.3 ± 0.1	3.5 ± 0.1
	10	4.0	5.8	--	--	--
	20	4.3	5.8	--	--	--
	30	5.7	5.9	--	--	--
HPP-200 MPa	0	1.8	1.2	3.3 ± 0.2	13.0 ± 0.1	3.5 ± 0.1
	10	--	--	--	--	--
	20	--	--	--	--	--
HPP-300 MPa	0	ND	ND	3.9 ± 0.3	12.7 ± 0.2	3.5 ± 0.1
	10	ND	ND	--	--	--
	20	ND	ND	--	--	--
	30	2.2	3.2	--	--	--
HPP-400 MPa	0	ND	ND	4.3 ± 0.4	12.2 ± 0.1	3.7 ± 0.1
	10	ND	ND	--	--	--
	20	ND	ND	--	--	--
	30	ND	ND	--	--	--
HPP-500 MPa	0	ND	ND	4.4 ± 0.2	11.9 ± 0.1	3.7 ± 0.1
	10	ND	ND	--	--	--
	20	ND	ND	--	--	--
	30	ND	ND	--	--	--

ND= no detectable survivor. -- Not evaluated. The limits of detection were <1.0 log CFU/g.

Conclusions

HPP at 400 MPa can be used as a potential alternative for minimal processing that can guarantee the safe consumption of *Reineta* apple puree. Moreover, this work provides inputs on the use of this technology in other food matrices.

References

- FAOSTAT. (2022). *Crops*. Food and Agriculture Organization of the United Nations.
- FDA. (2004). Food and Drug Administration, Center for Food Safety and Applied Nutrition. FDA-2002-D-0298
- Landl, A... Effect of high pressure processing on the quality of acidified Granny Smith apple purée product. *Innovative Food Science and Emerging Technologies*, 11(4), 557–564.
- Pino-Hernández, E... Hydrothermal and high-pressure processing of chestnuts - Dependence on the storage conditions. *Postharvest Biology and Technology*, 185, 111773.
- Pinto, C. A... Effects of high-pressure processing on fungi spores: Factors affecting spore germination and inactivation and impact on ultrastructure. *Comprehensive Reviews in Food Science and Food Safety*, 19(2), 553–573.

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