Shelf-life extension of watermelon juice preserved by hyperbaric storage at room temperature compared to refrigeration

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Keywords: Hyperbaric storage, Refrigeration, Shelf-life, Watermelon juice

Abstract

This work reports Hyperbaric Storage (HS) preservation of raw watermelon juice at variable/uncontrolled room temperature (RT, z 21 °C) for 7 days at 100 MPa and compared it with refrigeration (RF). At the end of storage, there was an increase in microbial counts (total aerobic mesophiles, psychrophiles, and yeasts and moulds) to above 6 log10 CFU/mL for samples stored at atmospheric pressure (RFand RT), while juice stored under HS/RT showed maximum values of about 2 log10 CFU/mL for total aerobic mesophiles/ psychrophiles and below the detection limit for yeasts and moulds. HS/RT juice showed also physico- chemical parameters at levels similar to the initial juice. Thus, HS/RT can not only be used to preserve foods with no refrigeration energetic costs (since it does not require temperature control), but additionally, has also a great potential to extend the shelf-life of food products, compared to RF. This is the first case in the literature showing this additional potential/advantage of HS/RT.

1. Introduction

High pressure technology has assumed a key role on non- thermal food pasteurization of fruit juices, vegetables, meat, and seafood (Bermúdez-Aguirre & Barbosa-Canovas, 2011). Notwithstanding, a new methodology for food preservation using pressure (Hyperbaric Storage, HS) was proposed to store food products under pressure at uncontrolled/variable room temperature (RT), with high potential energy savings, since energy is only required to compress and decompress, while refrigeration (RF) needs constant power supply (Fernandes et al., 2014). It is estimated that more than 50% of the energy spent on food industries is related with RF facilities (James & James, 2010; Tassou, Lewis, Ge, Hadawey, & Chaer, 2010).

Lately, research on HS/RT has increased to deepen the knowledge about the feasibility and potential of HS to preserve different food products with distinct characteristics. The few studies concerning HS/RT of highly perishable food products (non-acidic and high water activity) were carried out using watermelon juice (Fidalgo et al., 2014; Santos et al., 2015), melon juice (Queiros et al., 2014), Requeijão (a Portuguese whey cheese) (Duarte et al., 2014), carrot soup (Moreira et al., 2015), and sliced cooked ham (Fernandes et al., 2015), showing, generally, that HS/RT performed equal to better than RF regarding microbial growth inhibition and also in what concerns to maintain basic intrinsic physicochemical quality parameters. Nevertheless, these works only describe HS experiments for short time periods (up to 60 h) (Fidalgo et al., 2014). In another work (Segovia-Bravo, Guignon, Bermejo-Prada, Sanz, & Otero, 2012), it was studied strawberry juice stored under pressure over 15 days. However, this juice is an acidic food product (pH z 3.3), what poses a natural hurdle to microbial growth and thus a higher shelf-life. For the study just cited, at the end of the 15 days, both refrigerated and HS stored strawberry juices were still acceptable for consumption.

Watermelon juice is a highly perishable food product due to its low acidity and high water content, presenting a shelf-life of z 4 days (Fidalgo et al., 2014; Santos et al., 2015) under RF. Therefore, the aim of this study was to carry out a shelf-life evaluation and comparison of raw watermelon juice preserved by HS (100 MPa) at naturally/variable RT (z 21 °C) and RF in a 7 days study. Microbiological (total aerobic mesophiles, total aerobic psychrophiles, and yeasts and moulds) and physicochemical parameters (pH, total soluble solids, cloudiness, browning degree, and colour) were analysed and compared to samples stored under RF for the same time period. Apressure of 100 MPawas selected for this study, since in previous studies on HS this level was very effective to slow down to be clearly longer than the usual shelf-life of watermelon juice by RF (around 4 days).

2. Materials and methods

2.1. Juice preparation and storage conditions

Seeded red watermelon (Citrullus lanatus) was purchased at a local supermarket and kept at 4 °C until washing, peeling, crushing, and filtration with a sterilized cotton filter to separate seeds. In sterile conditions, using a laminar flow cabinet (BioSafety Cabinet Telstar Bio II Advance, Terrassa, Spain) to avoid contaminations, the juice was aseptically placed in low permeability polyamide/poly- ethylene bags (PA/PE-90, Albipack-Packaging Solutions, Agueda, Portugal) previously sterilized with UV light.

HS was carried out at 100 MPa for 7 days at uncontrolled/variable room temperature (RT, z 21 °C) using a 2-L high pressure equipment (FPG7100, Stanstead Fluid Power, Stanstead, United Kingdom). At the same

time, two control samples were kept at atmospheric pressure at RT (AP/RT) and refrigeration (RF), submersed in the same pressurization fluid and kept in the dark to keep samples in the same conditions (except for pressure).

2.2. Microbiological analyses

Total aerobic mesophiles (TAM) and total aerobic psychrophiles (TAP) were enumerated in plate count agar (Merck, Germany), by incubation at 30 ± 1 °C and 20 ± 1 °C for 3 and 5 days, respectively (ISO 4833: 2003). Yeasts and moulds (YM) were plated for counting in rose bengal chloramphenicol agar (Merck, Germany), and incubated at 25 ± 1 °C for 5 days (ISO 7984: 1987). Samples for microbiological analyses were obtained in duplicate, each one plated in triplicate and the results were expressed as log10 colony forming units per millilitre of watermelon juice (log10 CFU/mL).

2.3. Physicochemical parameters

The pH value was determined at 25 ° C using a properly calibrated glass electrode (pH electrode 50 14, Crison Instruments, S.A., Spain). The total soluble solids content was measured using the method described by Wang et al. (2005), expressing the results in °Brix. For browning degree (BD) determination, juice was centrifuged at 9000 x g, at 4 °C for 20 min and the supernatant absorbance was measured at 420 nm in a UV-VIS spectrophotometer (Microplate Spectrophotometer Multiskan Go, Thermo Scientific, USA), following the method used by Zhang et al. (2011). The cloudiness values were obtained by direct measurement of absorbance (700 nm) in the aforementioned spectrophotometer, using the method indicated by Singleton & Rossi Jr., (1965).

Juice colour was assessed using the following colour parameters: red/green colour (a^*), yellow/blue colour (b^*), and luminosity (L^*) parameters. The total colour variation (DE^*) was calculated ac- cording to the equation (1) (Liu, Hu, Zhao, & Song, 2012):

$$\Delta E^* = \left[(L^* - L_0^*)^2 + (a^* - a_0^*) + (b^* - b_0^*) \right]^{1/2} \quad (1)$$

where DE^* is the total colour variation between the sample (juice after storage) and the control (initial juice values are identified by the subscript 0).

Each parameter was obtained from duplicated samples, each one measured in triplicate.

3. Results and discussion

3.1. Microbiological analyses

According to Fig. 1, HS/RT was effective not only in maintaining TAM and TAP, but even in decreasing them, since microbial counts were reduced by almost 2 log10 CFU/mL compared to the initial value. Regarding YM counts, juice stored at HS/RT presented values below the detection limit (1 log10 CFU/mL) after 7 days of storage, showing also a reduction of at least 2.6 log10 CFU/mL.

Contrarily, samples stored under RF and AP/RT showed an in- crease of TAM and TAP values of at least 2 log10 CFU/mL compared to the initial value, with values after 7 days being above the acceptance limit (6 log10 CFU/mL) for all microorganisms analysed (TAM, TAP and YM).

Additionally, it was possible to verify informally and qualitatively that juice samples stored at HS/RT showed an odour and freshness similar to the initial one, while juice samples stored at RF and AP/RT showed a clear unpleasant odour at the 7th day of storage.

Results previously obtained by Santos et al. (2015) and Fidalgo et al. (2014) already indicated watermelon juice microbial stability under HS/RT during 8 and 60 h (2.5 days), respectively, but experiments over longer storage periods were not carried out before. So, in this short communication, the results indicated that HS/RT at 100 MPa allowed a clear microbial stability of this juice over, at least, 7 days of storage, compared to RF, resulting in a longer shelf-life due to microbial inhibition and additional microbial inactivation compared to the initial juice.

3.2. Physicochemical parameters

The pH values were stable during HS/RT (5.92), being very similar to RF (5.80) and that of the initial juice (6.17), while AP/RT showed a higher pH variation, reaching 4.79 (Table 1), which is correlated to the higher microbial loads of these samples.

Concerning total soluble solids content, HS/RT and RF showed similar values (8.0 °Brix) to the initial juice (8.1 °Brix), while for storage at AP/RT it showed a considerable decrease (to 6.9 °Brix), which might be related with sugars consumption by microorganisms, since a high microbial development was observed for those samples.

Regarding BD, the values HS/RT and RF samples (0.22), were

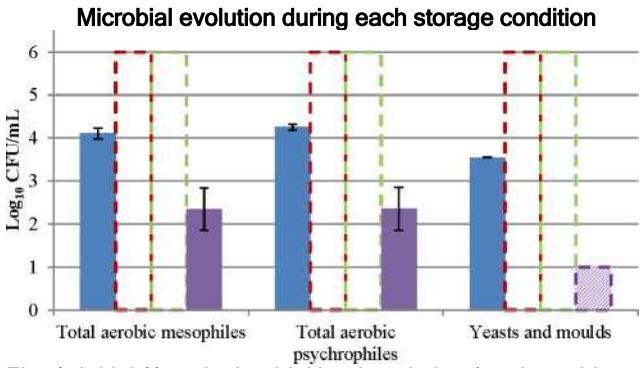

Fig. 1. Initial ( ) and microbial load evolution (total aerobic mesophiles, total aerobic psychrophiles and yeasts and moulds) over 7 days of storage, in the three conditions studied: AP/RT (storage at atmospheric pressure and room temperature, = 21 °C – ■), RF (storage under refrigeration, 4 °C – ■) and HS/RT (hyperbaric storage at room temperature, = 21 °C – ■). Discontinuous line bars indicate values above 6 $\log_w$ CFU/ mL (open bars) or below 1 log10 CFU/mL (filled bar).

Table 1
Effect of different storage conditions on the physicochemical parameters of watermelon juice (±standard deviation).

Storage time (days)	Storage conditions	pH	°Brix	Browning degree	Cloudiness	Colour			
						L^*	a^*	b^*	ΔE^*
0	Initial juice	6.17 ± 0.07	8.1 ± 0.1	0.26 ± 0.04	0.64 ± 0.06	33.88 ± 0.59	11.87 ± 0.42	7.33 ± 0.44	–
7	AP/RT [a]	4.79 ± 0.01	6.9 ± 0.1	0.10 ± 0.01	1.21 ± 0.02	35.01 ± 0.14	10.30 ± 0.39	5.29 ± 0.16	2.52 ± 0.34
	RF [b]	5.80 ± 0.04	8.0 ± 0.1	0.22 ± 0.01	0.67 ± 0.01	35.12 ± 0.40	11.38 ± 0.36	5.39 ± 0.29	2.06 ± 0.13
	HS/RT [c]	5.92 ± 0.14	8.0 ± 0.1	0.22 ± 0.03	0.51 ± 0.01	36.38 ± 0.54	10.46 ± 0.48	5.98 ± 0.39	3.18 ± 0.31

[a] Storage at atmospheric pressure and room temperature (AP/RT, ≈ 21 °C).
[b] Storage at atmospheric pressure and refrigeration (RF, 4 °C).
[c] Hyperbaric storage at room temperature (HS/RT, 100 MPa and ≈ 21 °C).

similar to initial value (0.26), while a decrease of more than 50% was verified for AP/RT samples (0.10). The decrease of BD AP/RT samples might be due to the heavy microbial spoilage of these samples. Similar results were reported by Fidalgo et al. (2014), who verified a decrease of BD of samples stored at AP/RT for over 60 h, while HS/RT allowed a maintenance of samples BD.

Cloudiness showed a small decrease (about 10%) for HS/RT samples, presenting better results than the initial (from 0.64 to 0.51), while for RF samples the value was similar to the initial (0.67). For AP/RT condition it was verified an evident increase of cloudiness value of about 2-fold (1.21), which is possibly related with the activity of enzymes, such as pectin methylesterase and polygalacturonase (Bermejo-Prada, Segovia-Bravo, Guignon, & Otero, 2015; Fidalgo et al., 2014; Segovia-Bravo et al., 2012).

The juice total colour variation was slightly more noticeable for HS/RT samples (3.18), due to the increase of the L* parameter (about 10% compared to the initial value) compared to RF (2.06) and AP/RT (2.52).

In summary, HS/RT preservation resulted in similar or even better values than the ones observed in RF and compared to the initial values, with the exception of cloudiness and L* value.

4. Conclusions

Considering the results obtained in this work, HS at 100 MPa at naturally variable/uncontrolled RT (z 21 °C) for 7 days resulted in a juice with low microbial loads, while RF storage yielded a juice spoiled and unacceptable for consumption. These results indicate a clear self-life extension of the juice preserved by HS/RT, compared to RF and additionally a microbial reduction compared to the initial juice.

HS/RT preservation resulted in physicochemical parameters similar to RF, except for colour, where was observed a slight in- crease of DE* (caused mainly by the L* value increase) and a slight higher decrease for cloudiness.

These are the first results in literature comparing the shelf-life of a food preserved by HS/RT with RF, for a period overpassing the shelf-life achieved by RF. The lower microbial loads verified for preservation by HS/RT point even for a shelf-life longer than 7 days. More research is required to further expand the knowledge about HS/RT full and the real potential to preserve food products, namely longer storage periods, pathogens behaviour, and sensorial/organoleptic characteristics.

Acknowledgement

Thanks are due to FCT/MEC for the financial support to the QOPNA research Unit (FCT UID/QUI/00062/2013), through national funds and where applicable co-financed by the FEDER, within the PT2020 Partnership Agreement.

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This is a post-peer-review version of an article published in LWT - Food Science and Technology, 2016 following peer review. The version of record Pinto, C., Moreira, S., Fidalgo, L., Santos, M., Delgadillo, I., Saraiva, J. (2016). Shelf-life extension of watermelon juice preserved by hyperbaric storage at room temperature compared to refrigeration. LWT - Food Science and Technology, 72, 78-80. https://doi.org/10.1016/j.lwt.2016.04.036