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Production of low-fat yogurt with quince (Cydonia oblonga Mill.) scalding water

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ABSTRACT

Quince scalding water is rich in phenolic compounds and flavonoids which provide interesting antioxidant properties, and also contain organic acids and sugars. The aim of this study was to evaluate the direct use of quince scalding water for set style yogurt production. The addition of quince scalding water provided color changes and reduced yogurt sensory scores. Quince scalding water had inhibitory effect against lactic acid bacteria, probably due to its high content in polyphenols. As a consequence, quince scalding water enriched yogurts had higher pH and lower lactic acid content compared to control yogurts. Such changes are reflected in their rheological and textural properties: soft yogurts of higher deformability and lower elastic behavior and viscosity. Future research on the addition of quince scalding water to other foods, or the study of their antibacterial or antioxidant properties would be of great interest.

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1. Introduction

Waste water could result from several steps in the food industry like the cleaning process, the step of blanching fruits or vegetables, or as secondary by-product such as washing water during the extraction of dietary fiber. The possibility of successfully including these waste water by-products in the human food industry would help in enhancing the economic development quince producers and processors.

Quince fruit (*Cydonia oblonga* Miller) is not appreciated fresh because of pulp hardness, bitterness and astringency. But when ripe it is highly demanded for processing 'marmalade', jams, jelly and cakes (Silva et al., 2002; Silva et al., 2005). Rodríguez-Guisado et al. (2009) determined the average values for the chemical characterization of five quince fruits collected in Southeastern Spain showing high water content (76.72 g/100 g), high crude fiber content (5.33 g/100 g), and low-fat content (1.95 g/100 g). Quince (like apple and pear) is classified into the Roseaceae family and their well-established beneficial properties to human health were found mainly related to their phenolic content. Fattouch et al. (2008) reported that quince pulp extract showed a superior phenolic content (66.95 mg/100 g fresh weight) than apple pulp (27.44 mg/100 g fresh weight) and pear (24.38 mg/100 g fresh weight). So the fact that quince fruits are a source of sugars (mainly

fructose, glucose, and sucrose) and phenolic compounds with antioxidant activity suggests that it is likely that a small portion of these compounds become part of the scalding water when they are processed. Quince healthy properties have been explored by several authors, a recent study has proposed the use of quince hot water, scalding water, as a functional food for its anti-allergic effects on type I allergic symptoms proved in mice and *in vitro* cells (Shinomiya, Hamauzu, & Kawahara, 2009). However, the reuse of water in the food industry has limits, as has been reviewed by Casani, Rouhany, and Knøchel (2005). The type of water that we are using in the present study is classified as water for direct recycling for non-food uses and cleaning (Casani et al., 2005), however, given the interesting compounds that contains and the references to its health benefits we considered to present a simple approach by directly using such water in foods.

Fermented milk products already have a positive healthy image due to the beneficial action of its viable bacteria and yogurt already have a record as being healthful (Heller, 2001). The addition of antioxidant food ingredients such as green tea and lemon, strawberry pulp and vitamin E was been also tested on dairy products (Jiménez, Murcia, Parras, & Martínez-Tomé, 2008). The purpose of incorporating ingredients known to have antioxidant activity is to increase the functionality and antioxidant activity of these foodstuffs and in this way to improve consumer's protection against pathologies related with free radicals (Jiménez et al., 2008).

The aim of this study was to evaluate the direct use of scalding water from quince fruits scalding in a set style yogurt on yogurt quality during 28 days of refrigerated storage.

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2. Materials and methods

2.1. Materials

Quince fruits were directly collected from the quince grove of the Miguel Hernandez University (Orihuela, Alicante, Spain). Quince fruits were separately scalded at 100 °C for 3 min and immediately divided into solids (seeds, pulp, peels) and liquid. For scalding, quince:water ratio was 1:1. Seeds and peels were handy removed, the flesh part was crushed to obtain a paste. Scalding water was filtered through a cotton gauze and frozen stored until use. For best knowledge the scalding water (quince scalding water, QSW) and the resulting quince fruit paste (QFP) were analyzed to compare how many bioactive compounds in the pulp and skin were released to the scalding water.

For all the study the same batch of skim milk powder was used (34 g/100 g protein, 52 g/100 g lactose, 1 g/100 g fat, 6.8 g/100 g ash, 5.2 g/100 g moisture; Central Lechera Asturiana, CAPSA, Granada-Siero, Spain). Commercial starter cultures of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* (Ezal® MY900, Rhodia Food-Danisco A/S, Sassenage, France) were used at the concentrations prescribed by the suppliers. Only for batches prepared for sensory analysis 8 g/100 g of sucrose was added to each mix (prior to pasteurization) to sweeten the yogurt.

2.2. Yogurt manufacture

Control and quince yogurt were manufactured. Skim milk powder was reconstituted with deionised water which is a common industrial practice (Augustin, Clarke, & Craven, 2003) and quince scalding water (QSW) at 15 g/100 mL for control and quince yogurt respectively. Reconstituted skim milk (RSM) was pasteurized in a water bath at 80 °C for 30 min, followed by immersion in ice-water baths to cool down to 43 °C, at this point the starter culture was added and gently shaken. The inoculated mix was poured into brand new 100 mL cups of yogurt and incubated at 43 °C to reach pH 4.7, and then cooled down to 4 °C.

2.3. Raw material analysis

2.3.1. pH and °Brix

pH and soluble solids (°Brix) were determined at 20 °C using a pH-meter and a refractometer respectively.

2.3.2. Chemicals and extraction process

1,1-Diphenyl-2-picrylhydrazyl (DPPH), ferrozine, Folin-Ciocalteu's reagent, gallic acid, iron (III) chloride, iron (II) chloride, trichloroacetic acid (TCA) and Trolox were from Sigma Chemical Company (Germany). Dibasic potassium phosphate, sodium carbonate and dibasic sodium phosphate were from Merck (Darmstadt, Germany). Potassium hexacyanoferrate was from Fluka BioChemika (Germany). The solvent used for the extraction process was methanol of HPLC ultra-gradient grade, supplied by Merck.

For the extraction process two hundred milligrams of quince fruit paste (QFP) were extracted for 2 h with 2 mL of methanol/ H_2O (50:50 v/v) at room temperature on an orbital shaker set at 600 rpm. The mixture was centrifuged at 1000g for 15 min, and the supernatant was decanted. Quince scalding water was directly used for analysis.

2.3.3. Measurements of total phenol content

The total phenol content (TPC) was determined using Folin-Ciocalteu's reagent (Viuda-Martos, Ruiz Navajas, Sánchez Zapata & Fernández-López, 2010).

2.3.4. Measurements of total flavonoid content

For total flavonoid content (TFC), the method based on Blasa et al. (2005).

2.3.5. Determination of antioxidant activity

2.3.5.1. 2,2'-Diphenyl-1-picrylhydrazyl radical scavenging method. 2,2'-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method was run according to Viuda-Martos et al. (2010)

2.3.5.2. Ferric reducing antioxidant power. Ferric reducing antioxidant power (FRAP) of the QFP extracts (100 g/L) or scalding waters was determined by using the potassium ferricyanide-ferric chloride method (Oyaizu, 1986).

2.3.5.3. Ferrous ion-chelating ability assay. Ferrous ion-chelating ability assay (FIC) assay was carried out according to the method of Singh and Rajini (2004).

2.3.6. Organic acids and sugars analysis

The content of organic acids and sugars were determined as follows: 5 g samples with 10 mL ultrapure water acidified with 0.1 g/ 100 g phosphoric acid H₃PO₄ were homogenized for 20 s at 13500 rpm and centrifuged for 20 min at 15000 rpm. Triplicate extractions were obtained from each sample. The supernatants fluids were filtered through 0.45 µm membrane filter (Millipore). Ten microliter samples were injected in a cation exchange column (Supelcogel C-610 H, 300×7.8 mm, Supelco, Bellefonte) with a precolumn (Supelguard-H. 50×4.6 mm, Supelco, Bellefonte), using $0.1 \text{ g}/100 \text{ g H}_3\text{PO}_4$ as mobile phase, operating flow rate of 0.5 mL/min. A Hewlett Packard HP-1100 instrument (Woldbronn, Germany) coupled with two detectors: DAD G1315A (set at 210 nm) and RID G-1362 A was used. Standards of organic acids, monosaccharides and oligosaccharides were obtained from Supelco. Samples were run at 30 °C and the run time was 30 min (Doughty, 1995). Peaks were identified by comparison with retention time of the standards, and quantified by regression formula obtained with the standards.

2.4. Yogurt analysis

2.4.1. Physicochemical analysis

2.4.1.1. Color measurements. The CIE LAB color space of yogurts was studied. Color determinations were made at 12 ± 2 °C by means of a Minolta CM-2002 (Minolta Camera Co., Osaka, Japan) spectrophotometer, with a liquid accessory CR-A70 (Minolta Camera Co., Osaka, Japan), with illuminant D_{65} and an observer of 10° . Six measures per sample were taken.

2.4.1.2. Texture analysis. Penetration test was performed with a Texture Analyser TA-XT2 (Stable Micro Systems, Surrey, England) and a 5-kg load cell was used. Constant speed penetration tests were performed directly on cylindrical containers (4.5 cm diameter \times 4 cm height). All instrumental texture analyses were conducted at 7 °C. This is a 'destructive' test as no structure recovery is allowed. A cylindrical probe 10 mm diameter ebonite (P-10) was introduced 15 mm into the samples at a speed of 1 mm/s. From the force-versus-time curves, values for the maximum force (N) were calculated as force at a distance of 15 mm ($F_{\rm max}$). Triplicate measures for each yogurt were performed.

2.4.2. Rheological measurements

The rheological parameters were measured by oscillatory testing, in triplicate at 7 °C by means of a rheometer Rheostress 600 (Haake, Karlsruhe, Germany). Tests were performed on slices of 1.2 \pm 0.2 mm thickness and 35 mm diameter obtained using a sharp blade. It was verified that all samples were within the range of linear

viscosity. The geometry used was plate and plate with serrated platens (35 mm diameter, 1 mm gap). The measurements were conducted in triplicate. Frequency test was run: frequency sweep from 0.01 to 10 Hz at a constant stress of 0.4 Pa with three cycles of measurements in both tests.

2.4.3. Sensory evaluation

Panelists (36) were members of the staff and students of the Miguel Hernández University, Alicante, Spain. All sensory work was carried out in the sensory laboratory at the University, which fulfills requirements according to the International Standards (ISO, 1988). In all cases a panel of three experts defined the attributes to be evaluated in yogurts. Panelists accepted to taste the samples before the tests, and they were informed of the type of product being tested and asked about yogurt consumption habits. Tap water was provided between samples to cleanse the palate. White plastic cups with 40 mL of yogurt at 10 °C were provided. Two samples were evaluated in each session. A total of three sessions were run. A seven-point hedonic scale (from dislike extremely to like extremely) was used to rate the following parameters: appearance, creaminess, sweetness, astringency, sourness, flavor and overall acceptance.

2.4.4. Microbiological analysis

Microbial analyses of yogurts were run at days 1, 7, 14, 21 and 28 of cold storage. Streptococci were counted on M-17 agar (Scharlau, Barcelona, Spain) incubated aerobically at 37 °C for 24 h. Lactobacilli were enumerated using MRS agar (Cultimed, Panreac, Castellar del Vallés, Barcelona, Spain) under anaerobic incubation at 37 °C for 48 h. Counting of yeasts and moulds were done on Rose Bengal agar with Chloramphenicol (Scharlau, Barcelona, Spain) using the spread plate technique and plates were incubated under aerobic conditions at 28 °C for 5 days. Samples were analyzed in duplicate.

2.4.5. Organic acids and sugars analysis

Organic acids and sugars from yogurts were analyzed during cold storage, as previously described in Section2.3.6.

2.5. Statistical analysis

PASW Statistics 18 software package, SPSS Inc., IBM (Chicago, IL, USA) was used. General Linear Model procedure was tested to evaluate factors (water source and storage time effect on yogurt samples). Tukey's test was used for means comparison (95% confidence level). For sensory data of yogurts and scalding water samples one way anova test was used. The whole experiment was independently run in triplicate.

3. Results and discussion

3.1. Raw material characterization

3.1.1. pH and °Brix

The average pH was 3.92 for QFP. °Brix was 11.73 in QFP. Rodríguez-Guisado et al. (2009) reported a quince fruit pH ranging from 3.60 to 3.84 and total soluble solids (as °Brix) from 11.57 to 14.70. The pH of quince scalding water was 4.83 and the °Brix 3.53.

3.1.2. Organic acid and sugar profile in raw materials

Table 1 shows the profile of sugars and organic acids of quince paste and scalding water and reconstituted skim milk powder.

Fructose was the predominant sugar in either QFP or QSW followed by manitol and glucose in similar amounts. Main organic acid in QFP was acetic acid and in lower amounts formic, malic and succinic acids. QSW contained acetic and malic acids as predominant acids followed by tartaric, succinic and formic acids (Table 1). The

Table 1Organic acids (mg/100 g) and sugars (g/100 g) profile of reconstituted skimmed milk and quince paste and scalding water.

	RSM	Quince	
		Paste	QSW
Organic acids (mg/100 g)			
Oxalic acid	3.60 (0.44) ^d	N.d.	N.d.
Citric acid	233.41 ^c (24.3)	84.72 ^b (2.73)	34.41 ^{a,b} (5.42)
Tartaric acid	400.31 ^b (43.3)	88.68 ^a (1.04)	74.71 ^a (5.57)
Malic acid	$2.47^{a}(0.69)$	303.73 ^c (12.63)	97.81 ^b (28.13)
Ascorbic acid	$0.37^{a}(0.11)$	10.40 ^c (0.11)	2.89 ^b (1.14)
Succinic acid	N.d.	270.14 ^b (22.16)	63.69 ^a (24.42)
Lactic acid	122.23 (3.93)	N.d.	N.d.
Formic acid	N.d.	318.97 ^b (39.89)	53.92a (22.09)
Acetic acid	148.51 ^b (1.87)	684.60 ^c (49.29)	103.09a (34.24)
Fumaric acid	$3.36^{a}(0.12)$	11.01 ^b (0.99)	1.54 ^a (1.54)
TOTAL ACIDITY (g/100 g)	0.91 ^b (0.07)	1.77 ^c (0.12)	$0.43^{a}(0.12)$
Sugars (g/100 g)			
Sucrose	N.d.	0.23 ^b (0.00)	$0.06^{a}(0.01)$
Glucose	N.d.	2.12 ^b (0.08)	$0.60^{a}(0.15)$
Fructose	N.d.	5.09 ^b (0.18)	$1.42^{a}(0.36)$
Manitol	N.d.	2.23 ^b (0.07)	$0.62^{a}(0.15)$
Lactose	6.40 (0.65)	N.d.	N.d.
TOTAL SUGARS (g/100 g)	6.40 ^b (0.65)	9.67 ^c (0.33)	$2.70^{a}(0.67)$

RSM: reconstituted skimmed milk, QSW: quince scalding water; N.d., not detected. n=9.

Values with different superscript letters within the same line significantly differ (Tukey's test, *P < 0.05).

high presence of acetic acid was due to quince was over-ripened; organic acids provide information about maturation stage of the fruit. Rodríguez-Guisado et al. (2009) reported that the predominant sugars in quince fruit cultivated in Southeastern Spain are fructose and glucose (7.95 and 5.00 g/100 g, respectively) and malic the main organic acid (0.78 g/100 g) followed by tartaric acid (0.22 g/100 g) and acetic acid (0.13 g/100 g). Overall, QSW was able to extract a great part of organic acids (mainly tartaric acid) and sugars from quince fruit. In this line, several authors have successfully recuperated water-soluble polysaccharides presents in hot water extracts from different fruits (Mandal et al., 2009; Yang et al., 2009) and sucrose from the liquid content of the solid wastes of the citrus juice industry by reverse osmosis (García, Gozálvez, & Lora, 2002). On the other hand, QFP presented nearly 4-fold higher total acidity and total soluble solids than QSW.

Lactose was the main sugar in skim milk. Main organic acid identified in skim milk was tartaric acid followed by citric, acetic and lactic acids and small amounts of other organic acids (oxalic, fumaric, malic and ascorbic acids).

3.1.3. Total phenol and total flavonoid content

The total phenol and flavonoid contents of QFP and its scalding waters are presented in Table 2. As can be seen QFP contained higher amounts of phenolics and flavonoids than QSW. The presence of these compounds in scalding water is due to leaching during the scalding operation.

Quince (like apple and pear) is classified into the Roseaceae family and their well-established beneficial properties to human health were found mainly related to their phenolic content. Fattouch et al. (2008) reported that quince pulp aqueous acetone extract showed a superior phenolic content (66.95 mg/100 g fresh weight) than apple pulp (27.44 mg/100 g fresh weight) and pear (24.38 mg/100 g fresh weight). Furthermore, Silva et al. (2004) founded that antioxidant activity of quince pulp, peel and jam methanolic extracts were strongly correlated with TPC. Recently, Magalhaes et al. (2009) showed much higher contents of TPC (250 mg/100 g for a methanolic extract of quince pulp) than those of Fattouch et al. (2008).

^d Values in parentheses denote standard error.

Total phenolic content (TPC), total flavonoid content (TFC) and antioxidant activity of quince paste (QP) and its scalding water (QSW) at different concentrations (A = 2.5 g/100 g, B = 5 g/100 g, C = 7.5 g/100 g, D = 10 g/100 g).

Type of raw material	TPC (mg GAE ^e /L)	TFC (mg $RE^f/100$ g)	DPPH Inhibition	Inhibition (50 per cent)				FRAP TEAC ^g (mM Trolox/L)	M Trolox/L)		
			A	В	C	D	hIC ₅₀	A	В	C	D
QP QSW	$1595.67^{B} (7.27)^{i}$ $410.80^{A} (15.88)$	459.86 ^B (23.21) 70.40 ^A (2.90)	4.22^{aA} (0.61) 9.74^{aB} (0.23)	6.81 ^{a,bA} (0.61) 14.24 ^{aB} (0.14)	8.72 ^{bA} (0.08) 21.32 ^{bB} (2.07)	12.69 ^{cA} (1.00) 22.70 ^{bB} (0.05)	19.61 24.21	$0.07^{\mathrm{aA}} (0.00)$ $0.24^{\mathrm{aB}} (0.06)$	$0.10^{\mathrm{bA}} (0.00)$ $0.46^{\mathrm{bB}} (0.03)$	0.15 ^{cA} (0.00) 0.57 ^{bB} (0.02)	$0.19^{\mathrm{dA}} (0.01)$ $0.93^{\mathrm{cB}} (0.04)$

Values with different small superscript letters within the same line significantly differ (Tukey's test, *P < 0.05); Values with differnt capital superscript letters within the same column significantly differ (Tukey's test, *P < 0.05);

n=9. Callic acid equivalent.

Rutin equivalent. TEAC, Trolox equivalent antioxidant capacity.

 $^{-1}$ LC₅₀, concentration (g/100 g) for a 50 per cent. inhibition.

Values in parentheses denote standard error.

Different amounts of total phenolics have been reported in several food by-products such as juice by products from apple (46.00 mg GAE/g), strawberry (39.39 mg GAE/g) and pear (12.90 mg GAE/g), among others (Peschel et al., 2006). QSW contained higher levels of TPC than the reported in such juices, and the reported flavonoid content was also high.

3.1.4. Antioxidant activities

Table 2 shows the concentrations required to scavenge DPPH radical and the scavenging values as inhibition (50 per cent). QSW and QFP showed good radical scavenging effect. In the DPPH assay, the higher the antioxidant activity the lower the IC₅₀. Viuda-Martos et al. (2010) reported IC₅₀ values of 0.042 per cent and 0.053 per cent for ascorbic acid and butylated hydroxytoluene (BHT) as positive controls. Fattouch et al. (2008) reported that quince peels aqueous acetone extracts showed the greatest antioxidant effect on DPPH radicals (57 per cent of inhibition, corresponding to 0.44 mM trolox equivalents) and also quince pulp showed the strongest effect over pear, apple and quince pulps extracts analyzed.

Table 4 also shows the results of the FRAP assay. Quince paste, at all the concentrations analyzed, showed good ferric reducing capacity in terms of Trolox concentrations. The inhibition was concentration-dependent. Although both QSW and QFP showed antioxidant activity by DPPH and FRAP assays, no iron chelating ability was detected on quince paste or scalding water. Other water extracts from fruits like figs (Yang et al., 2009) and Doum palm fruit (Hsu, Coupar, & Ng, 2006) have demonstrated to have potent antioxidant activity.

3.2. Characterization of yogurt with quince scalding water

3.2.1. Acidification process

The enrichment of yogurts with QSW causes an initial pH reduction (P < 0.05) (Fig. 1) due to the acidity of the QSW. QSW provides a low pH together with high formic acid content (Table 1) that may stimulate the growth of lactobacilli (Robinson, 2003).

3.2.2. Physicochemical analysis of yogurts

3.2.2.1. Color. The addition of QSW had no effect (P > 0.05) on color coordinates L* (84.27 ± 1.11 for control yogurt and 83.07 ± 2.48 for quince yogurt) and b* (5.34 ± 0.16 for control yogurt and 6.15 ± 0.49 for quince yogurt), but significantly increased a* (to -2.74 ± 0.06 for control yogurt from -2.21 ± 0.23 for quince yogurt) as QSW had a light yellowish color.

3.2.2.2. Texture. QSW yogurts had significantly (P < 0.05) lower F_{max} values (20.52 ± 0.71 N) than control yogurt (32.56 ± 1.04 N),

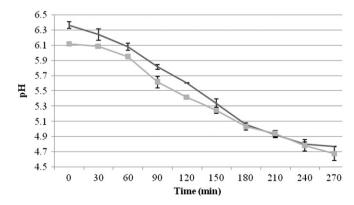


Fig. 1. Acidification curves of control yogurts () and quince enriched yogurts (). Data are the average $(n=9)\pm$ standard error (Tukey's test, *P<0.05).

even when QSW provided some soluble solids. In yogurt, the strength of the protein network increases along with the increase of lactic acid and exopolysaccharides released by lactic acid bacteria (Lubbers, Decourcelle, Vallet, & Guichard, 2004). So, the softening effect of QSW addition may have been due to the reduced populations of lactobacilli observed in QSW yogurts (Fig. 3b). QSW addition to skim milk powder induced a sudden pH decrease (Fig. 1) of 0.3 units which may have affected the casein structure during pasteurization and further set gel formation.

3.2.3. Rheology of set style yogurt enriched with quince scalding water

Oscillation tests allow the calculation of elastic modulus and loss modulus, G' and G'', which are related to energy stored and released, respectively, per deformation cycle, whereas $\tan (\delta)$ is associated with the degree of viscoelasticity of the sample (Singh & Muthukumarappan, 2008). Set style yogurts have flow properties that are characteristic of a non-Newtonian, weakly viscoelastic fluid with a highly time-dependent behavior (Ares, Paroli, & Harte, 2006). The studied yogurts remained in the linear viscoelastic region over the whole range of frequencies tested (0.01–1 Hz). G', G'', η^* , $\tan (\delta)$ and γ at 0.1 Hz were selected to run statistical analysis of the data (Table 3).

Coinciding with the observations of Sendra et al. (2010) the yogurts showed a predominantly elastic behavior (G' > G'') over the whole range of frequencies tested, which corresponds closely to that of a true gel. In general, moduli (G' and G'') increased with increased frequency. Our experimental values for the tangent of the phase shift of phase angle ($\tan \delta$) ranged from 0.210 to 0.246 which pointed to a concentrated amorphous polymer (Steffe, 1996, pp. 294–349) and a low value of $\tan (\delta)$ indicates that the gels had a predominant elastic character (more solid like). The linear decrease of the complex viscosity (data not shown) corresponds to a typical shear thinning profile.

The type of yogurt significantly affected rheological parameters (P < 0.05). G', G'', η^* decreased with the addition of QSW, whereas $\tan{(\delta)}$ and γ increased with its presence. Storage time only affected (P < 0.05) $\tan{(\delta)}$ and γ , both decreased with storage time, pointing to a hardening of the structure, probably related to the increase in lactic acid and exopolysaccharides released by lactic acid bacteria (Table 4). Under the same stress and frequency conditions, the strain (γ) achieved in QSW yogurts was higher than that of control yogurts, pointing to a softer structure as has been also detected by the penetration test.

3.2.4. Sensory analysis

There were significant differences in appearance, astringency, flavor and overall acceptability, which were best scored in control

yogurts than in QSW yogurts (Fig. 2). Creaminess, sweetness and sourness were adjudged to be very similar in all samples.

3.2.5. Evolution of pH and microbial counts during cold storage

Higher pH values were observed (P < 0.05) in QSW than in control yogurts. pH decreases gradually (Fig. 3a) during the storage period, presumably due to continued fermentation by the lactic acid bacteria (Dave & Shah, 1997).

Average initial microbial counts on yogurt samples were $\sim 10^8$ CFU/g. *S.thermophilus* was always present in higher numbers than *L.bulgaricus*. Counts of *S.thermophilus* and *L.bulgaricus* were significantly higher (P < 0.05) in control than in QSW yogurts (Fig. 3b), which is correlated with the lower pH of control yogurts. Numbers of *L.bulgaricus* decreased faster than did those of *S.thermophilus* which remained stable throughout the storage period. Previous studies have reported that the most important contributing factors for loss of cell viability are decreasing pH during product storage (post-acidification) and the accumulation of organic acids as a result of growth and fermentation (Kailasapathy, Harmstorf, & Phillips, 2008). Furthermore, QSW was already acidic and rich in organic acids although pH was not affected due to the buffering capacity of milk.

The presence of high amounts of formic acid and acetic acid may have accounted for the inhibitory effect of QSW on lactobacilli. In fact, Fattouch et al. (2008) reported antibacterial effect of quince against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Bacillus cereus*. Reported minimum inhibitory concentrations were quite low, from 10² to 10⁴ µg polyphenols/mL.

Certain yeasts play an important role in the spoilage of fermented products. Since milk is pasteurized before yogurt production, the presence of yeasts in yogurt is caused by recontamination processes during manufacture, and can be a problem in fruit-containing yogurts (Nogueira et al., 1998). Numbers of yeasts and moulds were significantly higher in QSW yogurts than in control yogurts (P < 0.05) and increased during cold storage, such increase may have been due to recontamination, however we recommend to pasteurize QSW prior to mix it with skim milk powder in order to enhance the microbial quality of the mix. The highest population was 5.00×10^2 CFU/g at day 28 in QSW yogurts.

3.2.6. Evolution of organic acids and sugar content in yogurts during cold storage

Evolution of organic acids and sugars of yogurts are presented in Tables 4 and 5 respectively. Organic acids are important indicators of bacterial metabolic activity in yogurt and they also contribute to the taste and flavor of the product along with other volatile and semi-volatile compounds such as diacetyl and acetaldehyde (Adhikari, Grün, Mustapha, & Fernando, 2002). All organic acids

Table 3 Evolution of rheological parameters of set yogurts enriched with quince scalding water under oscillatory testing (frequency sweep values obtained at 0.1 Hz, 0.4 Pa, 7 °C): G', elastic moduli; G'', viscous moduli; η^* , complex viscosity; $\tan(\delta)$ and γ deformation during 28 days of cold storage.

Type of yogurt	Storage	G' (Pa)		<i>G</i> " (Pa)		η* (Pa s)		tan δ $(-)$		γ ($-$)	
	time (days)	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.
Control	1	721.05 ^a	39.75	162.70a	7.20	1175.00 ^a	65.00	0.226 ^b	0.003	0.00054	0.00003
	7	753.70 ^a	153.71	163.20 ^a	35.21	1229.00 ^a	250.99	0.230^{b}	0.010	0.00057	0.00008
	14	1264.47 ^b	309.76	294.90 ^b	84.55	2066.67 ^b	511.25	0.215^{a}	0.005	0.00034	0.00013
	21	1160.33 ^b	216.71	245.27 ^b	44.49	1886.67 ^b	352.44	0.212^{a}	0.001	0.00036	0.00006
	28	1179.00 ^b	87.88	247.93 ^b	17.29	1916.67 ^b	141.11	0.210^{a}	0.003	0.00034	0.00026
Quince	1	417.80 ^a	43.18	102.70 ^a	7.89	685.00 ^a	63.24	0.246 ^b	0.002	0.00093 ^b	0.00001
	7	705.73 ^b	69.09	173.17 ^b	18.74	1156.67 ^b	112.60	0.245 ^b	0.005	0.00056^{a}	0.00005
	14	790.00 ^b	13.60	188.40 ^b	4.40	1295.00 ^b	25.00	0.239 ^{a,b}	0.002	0.00049^{a}	0.00001
	21	789.97 ^b	116.36	183.50 ^b	26.93	1293.00 ^b	191.29	0.232^{a}	0.002	0.00052^{a}	0.00008
	28	640.55 ^b	28.95	150.80 ^c	4.10	1045.00 ^b	45.00	0.236 ^a	0.005	0.00061a	0.00003

S.E., standard error.

Table 4 Evolution of oxalic, citric, tartaric, lactic, acetic, malic, ascorbic and fumaric acids (mg/100 g) of yogurts enriched with quince scalding water during 28 days of cold storage.

Organic acid (mg/100g)	Control yogurt					Quince scalding v	vater yogurt			
	1	7	14	21	28	1	7	14	21	28
Oxalic acid	1.49 ^b (0.08) ^d	1.49 ^b (0.06)	1.23 ^{a,b} (0.09)	1.09 ^a (0.10)	1.14 ^a (0.03)	3.54 (0.11)	3.11 (0.70)	1.62 (0.53)	1.57 (0.71)	2.82 (0.56)
Citric acid	198.94 (13.02)	183.21 (6.02)	177.96 (10.89)	171.21 (11.07)	187.65 (9.79)	257.82 (1.08)	274.69 (46.80)	180.73 (7.38)	182.52 (21.81)	249.17 (32.79)
Tartaric acid	265.5 (35.06)	221.82 (9.84)	225.58 (20.60)	229.4 (25.19)	244.48 (21.46)	303.5 (21.54)	363.32 (43.57)	259.52 (13.96)	249.64 (22.42)	335.03 (40.24)
Lactic acid	1192.56 (55.72)	1333.3 (54.73)	1294.17 (107.65)	1306.54 (98.79)	1383.85 (54.59)	1364.51 (89.65)	1612.28 (289.75)	1196.45 (24.01)	1318.41 (156.18)	1648.53 (179.96)
Acetic acid	11.13 ^a (0.39)	12.75 ^a (0.60)	13.33 ^{a,b} (0.88)	15.19 ^{b,c} (0.80)	16.01 ^c (1.12)	50.53 (4.95)	63.11 (12.53)	42.67 (1.21)	49.74 (6.25)	65.01 (6.76)
Malic acid	N.d.	N.d.	N.d.	N.d.	N.d.	7.49 (0.64)	4.96 (1.35)	9.15 (2.26)	5.69 (0.08)	8.87 (3.05)
Ascorbic acid	N.d.	N.d.	N.d.	N.d.	N.d.	0.84 (0.07)	0.88 (0.18)	0.78 (0.01)	0.69 (0.12)	1.33 (0.20)
Fumaric acid	N.d.	N.d.	N.d.	N.d.	N.d.	4.60° (0.52)	$2.33^{a,b}(0.47)$	3.12 ^{b,c} (0.19)	$1.07^{a}(0.06)$	$2.84^{a,b,c}$ (0.28)
TOTAL ACIDITY (g/100 g)	1.67 (0.14)	1.76 (0.02)	1.71 (0.21)	1.73 (0.19)	1.84 (0.17)	1.99 (0.07)	2.32 (0.40)	1.69 (0.05)	1.81 (0.21)	2.31 (0.26)

yogurts. No significant differences in the

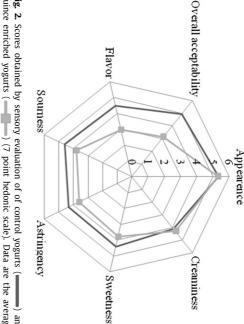
content of organic acids

N.d. = non detected.

Values with different superscript letters within the same line significantly differ (Tukey's test, *P < 0.05). No letters when no differences have been detected.

Fig. 3. Evolution of pH (A), and counts of lactobacilli (B) of control yogurts (= and quince enriched yogurts (=) stored at 4 °C for 28 days. Data average (n=9) \pm standard error (Tukey's test, *p < 0.05). Lactobacilli Log cfu/g) \Box pН 4.6 3.6 Time(days) Time (days) are the

detected were significantly (P < 0.05) affected by the type of yogurt (Table 4). QSW yogurts had slightly higher concentration of oxalic acid (P < 0.05) than control yogurts. Citric and tartaric acids were also present at similar concentrations but slightly lower (P < 0.05) **Fig. 2.** Scores obtained by sensory evaluation of of control yogurts (————) and quince enriched yogurts (————) (7 point hedonic scale). Data are the average (n=36) (Tukey's test, *P<0.05). Malic, fumaric and ascorbic acids were only detected in QSW produced from citrate, lactose and amino acids (Ong & Shah, 2009). at greater n the raw r Lactic acid was the major organic acid. Acetic levels in QSW yogurts Also, acid can be due to



^d Values in parentheses denote standard error.

evolution of maltohexaose and glucose (mg/100 g), galactose and lactose (g/100 g) of yogurts enriched with quince scalding water during 28 days of cold storage.

Type of yogurt	Storage time (days)	Maltohexaose (mg/100 g)	e:	Glucose (mg/100 g)	/100 g)	Galactose + Fructose (g/100 g)	Fructose	Lactose (g/100 g)	00 g)	TOTAL SUGARS (g/100 g)	ડર
		Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.
Control	1	84.66	3.43	N.d.		1.12	90.0	4.35	0.12	5.55	0.07
	7	76.61	1.82	N.d.	I	1.24	0.03	4.17	0.18	5.49	0.24
	14	72.67	7.12	N.d.	I	1.20	0.10	3.76	0.24	5.03	0.13
	21	72.21	7.26	N.d.	I	1.21	0.09	3.68	0.20	4.97	0.25
	28	83.07	2.48	N.d.	I	1.29	0.07	4.04	0.11	5.41	0.18
Quince	1	100.76	0.00	411.24	13.70	2.34	0.11	6.30	0.24	9.15	0.37
	7	102.77	19.24	416.94	84.25	2.58	0.52	6.29	1.42	9.38	
	14	72.82	3.87	296.12	5.26	1.85	0.04	4.31	0.10	6.53	
	21	72.36	12.96	291.93	29.64	1.95	0.22	4.27	0.50	6.58	
	28	92.64	12.70	406.44	48.62	2.54	0.31	5.77	08.0	8.81	1.17
d – non detected											ai.

were detected in yogurts during storage time except for lactic acid which increased with storage time (P < 0.05).

Lactose decreased with storage time (Table 5). Galactose increased or remained constant in all yogurts (P>0.05) which agreed with results reported by Wang et al. (2010). This is because of the cultures used in yogurt fermentation (L. bulgaricus and S. thermophilus) utilize the glucose moiety of lactose, but not the galactose moiety. Thus, while lactose and glucose content in fermented product decrease, the galactose content remains unchanged (O'Brien, 1999). Lactose content is much lower (P<0.05) in control yogurts than in QSW yogurts, which is correlated with the lower lactobacilli counts in QSW yogurts. Glucose was detected in QSW yogurts as it is provided by QSW and ranged from 0.3 to 0.4 g/100 g in yogurts.

4. Conclusions

Hot water extracts from different fruits are a good source of different compounds such as polysaccharides, sucrose, minerals, phenols and flavonoids, and many of them have antioxidant activity. Quince scalding water is rich in phenolic compounds and flavonoids which provide interesting antioxidant properties, and also contain organic acids and sugars that are all extracted during scalding. The addition of quince scalding waters provides color changes and reduced the sensory scores of yogurts due to its acidic nature. Such scalding water has inhibitory effect against lactobacilli, probably due to its high content in polyphenols. As a consequence, quince scalding water enriched yogurts have higher pH, lower lactic acid content and probably affected microbial metabolism for example reducing the release of exopolysaccharides from lactic acid bacteria compared to control yogurts. Such changes are reflected in their rheological and textural properties: softer yogurts of higher deformability and lower elastic behavior and viscosity. During cold storage of yogurts pH decreases, the gel structure is reinforced, lactobacilli population decreases (especially in quince enriched yogurts) and population of molds and yeasts increases. The direct use of heat treated QSW, although nowadays limited by regulations may enhance the eco-efficiency of quince industries. Further researches will be needed as well, to study the possibility of recuperate bioactive compounds from QSW that would allow their use as food ingredients (for their antibacterial or antioxidant properties) or as health promoting agents to ameliorate illness (such as allergy) or improve the nutritional value of conventional foods.

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