Research Article

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Settled and whipped yogurt enriched with calcium. Effect of two calcium salts on physicochemical and sensorial properties

Abstract

Yoghurt as one of the most important dairy products, is consumed worldwide, due to its nutritional and functional effects. Even though it's nutritional importance and richness, the incorporation of calcium contributes to augment its nutritional value. This research covered the incorporation of two calcium salts in both types of yoghurt, settled and whipped, on process and properties, calcium is an important nutrient for human, mainly for women.

Therefore, three commercial yogurts were characterized in order to identify their physicochemical nature, Lately, both types of yoghurt were elaborated, following recommendations of microorganism supplier and lab experience. And the largest part, in which settled and whipped yogurt were manufactured including two flavors and two calcium salts, measuring several properties, such acidity, color, density, pH, moisture, syneresis, total calcium, and viscosity. Acidity was higher as microorganisms grew in both types of yoghurt enriched with calcium; pH exhibited a neutralizing influence from calcium. Luminosity and redness as color parameters decreased with storage, as pH did; in contrast, the yellowness component augmented; without influence of the salt. Density, as well as moisture, did not show changes due to calcium enrichment. Syneresis resulted higher in whipped than settled yogurt. Apparent viscosity was higher in settled than in whipped systems, as expected; and both yogurts behaved as non-Newtonian liquids of pseudoplastic nature, in which flow properties were fitted to two rheological models. Sensory assessment, reflected influence of yoghurt type, flavor, salt type on acceptability, appearance, flavor, and texture.

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Paola Del Fabbro Macías,[1] Jorge F Vélez-Ruiz[1,2]

[1]Departamento de Ingeniería Química e Ingeniería en Alimentos Universidad de las Américas Puebla San Andrés Cholula, Mexcio
[2]Independent Consultant19 Oriente 1015, Mexcio

Correspondence: Dr. Jorge F Vélez-Ruiz, Universidad de las Américas Puebla, El Angel Puebla, Pue. Mexcio,
Tel +2222 370117

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Introduction

Yogurt as a milk product, is a very nutritive and demanded item. It contains proteins, minerals, fat, and lactose from milk, in which fat and lactose may be modified through manufacturing processes. There are a diversity of presentations and variants, maybe yogurt is one of the more novel and demanded food product; settled and whipped, fortified or not with minerals, with different fermentation degree, with less or more probiotics, defatted, lactose free or not. And even though the enormous variety of this dairy product, there are practical and scientific aspects that need to be researched. As it is known yogurt manufacturing implies milk standardization, pasteurization, fermentation with probiotic microorganisms, cooling and storage, as the more useful processing stages; thus, the manufacturing process may be modified in order to produce different yogurt presentations.

In addition to provide proteins of high biological quality, yogurt has many of the essential amino acids, among other benefits related with good health for consumers[1,2] analyzed the effect of fiber incorporation and fat decreasing on yogurt characteristics, physicochemical and flow, trying to improve the nutritive value of this milk product in both presentation types. In addition to acidity increasing, changes in color were observed, as well as pH decreasing, that were influenced significantly by the studied variables. Texture exhibited a significant modification through storage. Syneresis decreased with fiber incorporation and with fat decreasing; and as expected in flow response, the consistency coefficient augmented, and flow index decreased. Aportela-Palacios et al.[3] Also studied the rheological and physicochemical behavior of fortified (with fiber and calcium) of yogurt, in which they found an increasing in viscosity and a non-Newtonian behavior with fiber and calcium incorporation, also a decreasing in solids separation was observed. Two of these properties decreased with three weeks of storage, the consistency coefficient ranged from 0.32 to 2.0 Pa sn, and pH from 4.5 to 4.2, contrary to syneresis that augmented from 20 to 48%, and particularly as a function of yogurt system formulation.

Similarly, Vélez-Ruiz et al.[4] completed a work in which the influence of calcium and fiber on yogurt with low content of fat on properties, finding that consistency and syneresis were the most affected properties, calcium affected acidity, whereas fiber influenced pH and syneresis. The non-Newtonian nature of this item, varied through storage, as expected. More recently, Kilic et al.[5] investigated the re-production of lactic bacteria isolated from leguminous seeds in yogurt, studying bacteria viability, physicochemical and sensory responses. Similar properties, acidity, pH, syneresis, and sensory, between prepared and commercial yogurts were determined. These properties were notably affected by storage, and the isolated starter cultures of yogurt exhibited good potentiality as commercial ones. Sadighbathi et al.[6] studied the development and functional properties of low-fat yogurt enriched with postbiotic microorganism, on antioxidant activity, viability of starter cultures and quality parameters, finding improvements in the first, water holding capacity and sensory acceptability, this research was completed through 22 days.

There is not, doubt that yogurt is a very important dairy item, that covers a diversity of likes and necessities; and even though many investigations have been conducted on it, there are other aspects of science that should be researched, therefore this paper expresses those variables and results explored and obtained, of the incorporation of two sources of calcium on physicochemical and rheological properties of yogurt, in both, types of yogurt, settled and whipped.

Methodology

In order to reach the objectives of this work, several stages were completed: 1) commercial yogurt, without and with presence of this mineral, were analyzed; 2) yogurt was elaborated with the incorporation of two salts; 3) settled and whipped yogurts with two flavors (kiwi and passion fruit) were prepared, analyzing their properties; 4) a sensory assessment was developed on these studied yogurts. All analytical methods and procedures applied in this work, were in agreement with official and laboratory methodology.

Yogurt preparation

Yogurt was elaborated in labs of the IQA department of this University, following the next methodology: liquid milk was prepared by reconstitution of powder milk (Nido brand) acquired in a local supermarket, at 16% (w/v); two calcium salts were utilized, calcium mono and disodium phosphate, at levels of 250 and 300 mg in 100 mL of milk, and 125 and 150 mg in 100 mL of milk, respectively. Starter cultures were obtained with a specialized supplier, Wisby brand (Dantec. S.A. of CV) from Mexico city; a freeze-dried mix of Lactobacillus bulgaricus and Streptococcus thermophilus. (Jo-Mix-1-30 VibyvacR DIP). Kiwi and passion fruit flavors were acquired from Sodexin (Sabores y Perfumes, S.A of C.V.), also from Mexico city, specially developed for yogurt, and incorporated to both types.

Settled yogurt was manufactured, following the supplier recommendations; this is summarized next:

i) Reconstitution of liquid milk (16% w/v),

ii) Pasteurization of reconstituted milk, at 90°C during 30 min,

iii) Cooling down of pasteurized milk, to 42°C for starters inoculation,

iv) Addition of salts and incubation of starter microorganism, at 42° through 5 hours,

v) Cooling down to 20-25°C and addition of flavors, in which all systems were separated; and

vi) Refrigeration at 5 + 1°C.

Additionally, control yogurt was prepared without salt and flavor addition.

In case of whipped yogurt, the fifth step was different:

va) Cooling down to 10°C, and

vb) Agitation, with flavor addition, by using a mixer agitator (Omni GLH International) during 10 min.

Thus, the computed concentration of calcium in the correspondent yogurt were: 39.75 and 47.70 mg/100 mL, for monosodium phosphate with 250 and 300 (mg/100 mL), whereas 36.82 and 44.18 mg/100 mL, for disodium phosphate with 125 and 150 (mg/100 mL).

Microorganisms count

For microorganisms grown, samples of yogurt were taken each hour during five hours of fermentation through the manufacturing process, samples were preserved at refrigeration condition (5 + 1°C) and bacteria determination was completed at the next day, in according to the next method: agar MRS 8 (De Man, Rogosa and Sharpe), sterilized at 121°C-15 min, doing dilutions with sterile peptone water, 10-1 for the first incubation hour and 10-5 for the last hours; sowing in depth, followed by incubation, with inverted Petri dishes in a stove at 35°C through three days. A direct count in dish, determining the number of colonies per each dish, and considering the used dilution, allowed to convert the number of microorganism (UFC/mL) for each correspondent yogurt sample.

Physicochemical determinations

Acidity by titration with sodium hydroxide. Yogurt in continuous agitation was heated up to 37°C and sampled in an Erlenmeyer flask (5-18 g), in which distilled water and 0.5 mL of phenolphthalein were incorporated, then the hydroxide (0.1 N) was added to get a pink color .[7] One milliliter of hydroxide is equivalent to 0.009 g of lactic acid:

Acidity = 9 (mL of NaOH) (N of NaOH)/sample weight (Eq. 1)

Color with a Color Gard System 05 (Gardner Co.), using Hunter color parameters L for luminosity, a for red-green scale and b for yellow- blue scale.

Density by division of weights, between pycnometer with yogurt sample and pycnometer with water only.

Moisture by water evaporation, weighting an aluminum tray with sample, before and after 24 hours of heating at 100°C, and allowing cooling of the heated tray, in which:

Percentage of water: 100 (weight of tray with sample before heating/weight of tray after heating and cooling) (Eq. 2)

pH was measured with a Beckman potentiometer, by direct immersion of the electrode in the yogurt sample, and with previous standardization with buffer solutions of pH 4.0 and 7.0. Syneresis was determined by sample centrifugation, after 20 minutes [8] in which the percentage of solids separation resulted from next relationship: 100 (supernatant weight/ sample weight).

Calcium determination

This determination was completed with an Atomic Absorption Spectrophotometer (Varian Co). Yogurt samples were placed into lab stoves to dehydrate them through 1 day, after that, samples were incinerated in a muffle at 100°C; ashes were acidified with 2 mL of nitric acid and gauging to 100 mL with distilled water. From them, calcium solutions with 1, 2, 3, 4 and 5 ppm were prepared, adding strontium chloride (10 mL), potassium chloride (10 mL), and nitric acid (0.4 mL). Then, from prepared standards, and obtained lectures in the spectrophotometer, with flame of nitrous oxide-acetylene, using a wavelength of 422.7 nm and a bandwidth of 0.5 nm, to avoid chemical interferences; the calcium concentrations were quantified in milligrams per 100 milliliters of sample.

Viscosity evaluation

To measure the flow parameters of the different samples, a Brookfield viscometer model DVI was employed, with measuring spindles H1 and H2 for whipped yogurt, and spindles H3 and H4 for settled one; the sample was placed in a beaker of 400 mL in which a range of velocities of 0.5 to 100 rpm were applied; from this procedure, the lecture in torque percentage was also obtained. Then the application of three equations, to generate the shear rate and shear stress; the viscosities from slope of graphics or rheograms were obtained.[4,9] Thus, from shear rate and stress, and viscosity determinations, two flow models were applied to fit flow data:

$$\gamma = \frac{2\omega R_c^2}{(R_c^2 - R_b^2)} \qquad \text{(Eq. 3)}$$

$$\omega = \left(\frac{2\pi}{60}\right) N \qquad \text{(Eq. 4)}$$

$$\tau = \frac{M}{2\pi R_b^2 L} \quad \text{(Eq. 5)}$$

Where: γ is the shear rate (s-1), ω is the angular velocity of spindle (rad/s), Rc is the internal container radius (cm), Rb is the spindle radius (cm), N is the velocity of the spindle (rpm), τ is the shear stress (N/m2), M is the experimental torque (N·m), it is computed from a particular instrument relationship (6.737 x 10-5 multiplied by the instrument reading or lecture in percentage), and L is the effective length of the spindle (cm).

From previous information, two rheological models were used to fit data, flow models are expressed by equations 6 and 7, respectively: Power Law and Herschel and Bulkley fittings.[10]

$$\tau = K\gamma^n \quad \text{(Eq. 6)}$$

$$\tau = \tau_0 + K\gamma^n \quad \text{(Eq. 7)}$$

Where: τ is the shear stress (Pa), K is the consistency coefficient (Pa•sn), γ is the shear rate (s-1), n is the flow behavior index (dimensionless), and τo is the yield stress (Pa).

In order, to know the best fitting when both models were applied, a goodness relationship was applied. The percentage of mean relative error (PEM, Eq.) was computed as:

$$PEM = \frac{100}{n}\sum_{i=1}^{n}\left(\frac{\left|\tau_{\exp} - \tau_{pred}\right|}{\tau^{|}\exp}\right) \quad \text{(Eq. 8)}$$

Where: n is the number of measurements, τexp: is the shear stress (Pa), obtained experimentally and τpred: is the shear stress predicted by the applied model (Pa).

Sensory assessment

A sensory evaluation of the samples was performed by a panel of non-trained 7 tasters, who applied and hedonic scale of 9 points; characteristics as appearance, flavor, texture and global acceptability were qualified. A triangular test was also conducted, trying to recognize the yogurt with added calcium, based on their flavor.[11]

Results and discussion

Analysis of commercial yogurts

The physicochemical characteristics of two brands of whipped yogurt and three brands of settled one enriched with calcium salts, were completed, as a first stage of this work; strawberry (71 mg Ca/100 g) and nut with cereals (85 mg Ca/100 g) for whipped, whereas natural, pineapple-mango (267 mg Ca/100 g) and strawberry (115.6 mg Ca/100 g) for settled yogurts, also added with calcium (tricalcium phosphate) were analyzed, they were acquired in Puebla city.

Those selected and measured characteristics for commercial yogurt, are included in Table 1.

Acidity was influenced by calcium addition, attributed to Lactobacillus bulgaricus grown favored by salt, in agreement with Rivas and Vélez.[12] only one flavored sample had more than 1%, natural settled yogurt had the largest percentage (1.345). Color exhibited high luminosity, being the natural the most, the other color parameters varied as a function of the fruit/flavor. Density was similar in a range of 1041 to 1094 kg/m3, did not show effect of calcium; as well as moisture, with a range of 78 to 88%, and pH with a narrow range of 4.2 to 4,5, certainly being natural yogurt with 4.48, higher than others. Syneresis was lower in settled (28-33%) than in whipped (45-55%), in those added with fruits. With this information of those commercial yogurt characteristics, a panorama and context of them were considered, in order to complete our research with yogurt enriched with calcium.

Calcium determination testing

Determination of calcium at selected levels, were quantified by the spectrometric absorption method, those levels corresponded to added concentration, with next details: 300 mg of MPh/100 mL, was computed as 163.95 mg/100 mL and experimentally measured as 160 mg/100 mL (error 2.41%); whereas the other levels were next: 250 mg of MPh/100 mL, was computed as 156 mg/100 mL and experimentally measured as 136 mg/100 mL (error 12.8%); 150 mg of DPh/100 mL, was computed as 160.44 mg/100 mL and experimentally measured as 146.67 mg/100 mL (error 8.6%); and 125 mg of DPh/100 mL, was computed as 153.07 mg/100 mL and experimentally measured as 140 mg/100 mL (error 8.5%). The calcium concentration was also measured in control yogurt: computed as 116.25 mg/100 mL and experimentally measured as 108 mg/100 mL (error 7.1%). An overall approximation or accuracy of 92.2% between the measured and the computed was obtained, being very clear, as expected, than control had lower level with respect to enriched yogurts.

Influence of calcium on yogurt (exploratory analysis)

To know the effect of calcium phosphate, monobasic (MPh) and dibasic (DPh) on the elaboration process of both yogurt types, three aspects were analyzed, acidity, pH and microorganisms grown; the results were compared with a control yogurt (CY). The process was taken at time 0, when calcium salts were added, and the determinations were done each hour, during five hours, that is considered as the time to reach a pH optimum; in agreement with recommendations given by the supplier. Table 2 include the obtained results at this stage.

As expected, both parameters, acidity and microorganisms, augmented with process time, from 0.209 (at the beginning) to 1.027% (after 5 hours), and from 4.8 to 145 x105, respectively, for the control. Thus, the prepared yogurts exhibited acidities and microorganisms grown inside these values, with only exceptions of grown at the fifth hour. Those yogurts with DPh had lower acidity through the five hours of fermentation, than yogurts with MPh, in general. The microorganisms developing exhibited wide variations, but with the same increasing trend as a function of fermentation time. Obviously, pH was inversely proportional to the acidity. Then, with this information, we proceed to elaborate the yogurt systems to be studied at refrigeration storage.

Storage of prepared yogurts

For this part of the study, both yogurt types were prepared. The levels of calcium were similar to the exploratory part, and two flavors were added: kiwi and passion fruit; therefore, a total of 24 yogurt systems were analyzed through storage, 12 settled yogurts and 12 whipped yogurts: 5 with calcium salts and a control, for both flavors. Control yogurt was a yogurt without incorporation of calcium, just the flavor was added. Acidity and pH, inversely related, showed in general, an increasing and a decreasing trend, respectively, as expected.

The evolution of lactic acid through storage days is shown in Figure 1. Even though there was a general increasing, different trends are observed, in kiwi yogurt, acidity was lower than 1.3%, and the four systems with calcium showed a decrease in the seventh day, lately the increase is general, control had a decrease of this parameter at the last week. For passion fruit yogurt, acidity was lower than 1.6%, and a general increase was quantified for the first seven days, and a decrease was measured next days for systems with 150, 250 and 300 mg; whereas acidity in yogurt control and yogurt with 125 mg was constant after the seven day of storage. It is interesting how the added flavor influenced the acidity evolution. These levels of acidity are in according with the General Health Law in Mexico (1986) of 0.8 – 1.8%.

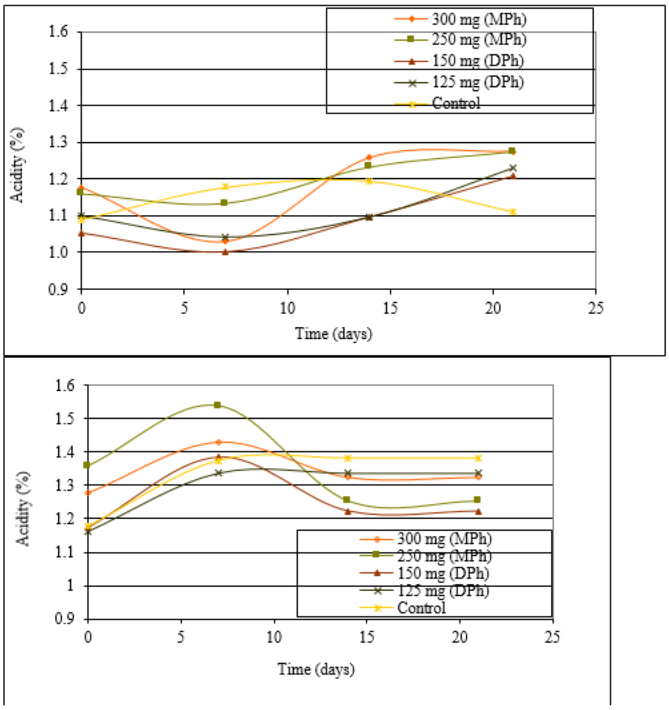

Figure 1 Acidity through 21 days of storage for settled yogurt, (a) kiwi flavor, (b) passion fruit flavor.

For whipped yogurt two acidity trends were observed, for kiwi flavor there was a small decreasing evolution, the acidity for day 0, was 1.22-1.32%; for day 7, was 1.20-1.32%; whereas for day 14, was 1.05-1.23%; and for day 21, was 1.20-1.21%. Passion fruit yogurt had a different response, 0.96-1.04%, 1.06-1.12%, 1.20-1.28% and 1.17-1.29%, for day 0, 7, 14 and 21, showing this last an increasing and expected trend. Control yogurts were consistent in acidity with yogurts enriched with calcium, exhibiting a decreasing in kiwi, 1.26-1.11% for settled, and 1.18-1.10% for whipped; in contrast an increasing in passion fruit yogurts; 1.18-1.39% for settled and 0.99-1.22% for whipped, were measured. In general, acidity showed significant effect of storage days and also, a significant effect was detected for flavor variable. Particularly, whipped yogurt was significantly influenced by calcium level.

pH decreased as expected, although yogurt systems presented, particular trends. At beginning settled, for kiwi:4.37-4.56 that changed to 4.28-4.36 at day 21, and for passion fruit, pH of 4.50-4.79 decreased to 4.30-4.63, correspondingly. Whipped yogurt varied from 4.47-4.82 at day 0, to 4.29-4.47 at day 21 for kiwi, and 4.44-4.73 to 4.29-4.51 correspondingly, for passion fruit. Control decreased pH, from 4.37-4.48 to 4.29-4.33 at the same storage time. And with exception of whipped yogurt of passion fruit flavor, the other three group of yogurts were significantly affected by salt level and storage time. Similar results were obtained by Vélez et al.[13] with calcium lactate and calcium chloride, as well as Pirkul et al.[14] with calcium lactate and gluconate, added to yogurt.

With respect to color, as other important physicochemical and quality property, three color parameters exhibited small changes through storage. Luminosity that was a little higher in yogurt with added calcium, showed a decrease from 88.0-89.5 at day 0, to 81.0-83.0 at day 21; a parameter augmented lightly (green scale), from –(2.0 - 3.0) at day 0, to – (1.9 – 2.5) at day 21, whereas b parameter exhibited a more no Table 1decrease (yellow scale), from 12.0-13.1 at day 0, to 7.2-8.7 at day 21; those lowest values corresponded to the control yogurts, in the three color parameters,. Taking these measures, net change of color, as representative of the three parameters are presented in Table 3, they are low, in general (< 2.4).

Table 1 Physicochemical properties (*) of five commercial yogurts

Yogurt brand	Acidity	Color			Density	Moisture	pH	Syneresis
	(% lactic acid)	Lh	ah	bh	(kg/m3)	(%)		(%)
Whipped with strawberry	0.768	68.35	16.63	9.19	1055.9	85.55	4.21	45.59
Whipped with nut and cereals	0.836	77.98	- 1.55	15.16	1063.9	81.7	4.35	54.45
Settled with pineapple-mango	1.227	60.49	1.25	40.26	1041.2	78.42	4.34	28.05
Settled with strawberry	0.865	69.37	13.62	5.56	1093.8	78.49	4.35	33.16
Settled natural	1.345	84.35	-3.29	9.94	1057	87.74	4.48	52.13

* MPh: calcium phosphate monobasic; DPh: calcium phosphate dibasic; Set: settled; Whi: whipped; p f: Passion fruit. Net change of color quantified with respect to corresponding yogurt controls.

As it is known the white color of yogurt is due to dispersion of light in which fat globules, colloidal particles and calcium phosphate are mainly involved; therefore, the incorporation of calcium salts increases the light reflection and white color. In general, ANOVA results, showed a significant effect of salt level on yogurt color for settled kiwi and whipped passion fruit; whereas storage time influenced significantly on both flavors in settled one. Part of color change in yogurt are attributed to loss of carotenoids and riboflavin by oxidation due to light exposure.[15] From computed values of net change of color, may be observed than changes were very low (< 2.4), indicating good stability of studied yogurts. For density, this property was lightly affected by calcium salt with respect to control, the density was constant (1010-1080 kg/m3), as expected, and did not show significant effect of storage time. Density of the studied systems is presented in Table 3.

Table 2 Evolution of parameters during previous analysis of the yogurt elaboration

Time (hours)		Acidity (%), pH and microorganism (CFU/mL x 105)			
	Yogurt with 250 and	300 mg (MPh)	125 and	150 mg of DPh	Control
0	0.437 6.56 3.70	0.44 6.57 3.72	0.23 6.56 4.80	0.251 6.56 4.01	0.209 6.56 4.80
1	0.482 6.07 4.37	0.493 5.99 4.25	0.26 6.29 4.01	0.261 6.06 4.80	0.235 6.42 5.70
2	0.51 5.82 3.56	0.543 5.74 3.90	0.273 6.17 2.49	0.298 6.14 3.61	0.347 6.26 3.69
3	0.715 5.39 5.37	0.73 65.56 4.19	0.51 5.89 8.96	0.543 6.01 9.94	0.507 5.75 8.05
4	0.856 5.00 23.0	0.888 5.08 10.7	0.854 4.75 33.4	802 4.83 15.5	0.866 4.83 65.4
5	0.926 4.89 346	0.931 4.98 259	0.915 4.56 233	0.932 4.53 563	1.027 4.56 145

Table 3 Physicochemical parameters for yogurt systems (*), at the beginning of the study

Yogurt system	Density (kg/m3)	Net change of color		Moisture (%)	Syneresis	
		Day 0	Day 21		Day 0	Day 21 (%)
300 mg MPh Set/kiwi	1044.6±20	1.56	2.44	85.12±0.08	47.67	41.67
250 mg MPh Set/kiwi	1045.9±10	1.14	1.49	84.44±0.03	46.98	41.15
300 mg MPh Set/p fruit	1056.0±20	1.23	1.72	84.60±0.06	44.92	43.73
250 mg MPh Set/p fruit	1054.9±30	0.74	1.39	84.43±0.02	40.57	42.14
300 mg MPh Whi/kiwi	1048.2±20	2.09	1.42	84.09±0.54	35.32	45.36
250 mg MPh Whi/kiwi	1048.0±20	1.58	0.95	84.56±0.46	39.82	38.5
300 mg MPh Whi/p fruit	1047.2±30	1.34	2.07	84.75±0.61	52.34	43.85
250 mg MPh Whi/p fruit	1046.3±10	1.44	2.08	84.54±0.51	52.44	46.04
150 mg DPh Set/kiwi	1042.5±10	0.99	2.34	84.50±0.23	41.39	34.17
125 mg DPh Set/kiwi	1043.5.±20	0.28	0.44	85.23±0.02	38.43	42.03
150 mg DPh Set/p fruit	1054.9±20	0.53	0.94	84.86±0.38	30.03	36.5
125 mg DPh Set/p fruit	1053.7±10	0.31	0.91	84.11±0.19	26.12	27.8
150 mg DPh Whi/kiwi	1048.6±40	1.14	0.62	84.75±0.14	33.6	42.35
125 mg DPh Whi/kiwi	1049.0±20	0.35	0.44	84.92±0.09	40.57	48.64
150 mg DPh Whi/p fruit	1048.7±20	0.64	0.33	85.16±0.03	49.15	43.85
125 mg DPh Whi/p fruit	1044.5±10	0.94	0.75	84.75±0.01	47.77	44.07
Control settled-kiwi	1041.7± 20	0	0	85.54±0.05	33.85	42.07
Control settled-p fruit	1053.4± 20	0	0	85.02±0.04	34.03	34.25
Control whipped-kiwi	1042.7± 10	0	0	85.41±0.66	44.93	36.13
Control whipped-p fruit	1041.8+ 20	0	0	85.57±0.02	46.43	42.33

Yogurts moisture exhibited little changes through storage, being higher in control. This property is also included in Table 3. As expected, yogurt control had higher moisture content (> 85.02%), due to the incorporation of calcium salts in the rest of the yogurt systems (84.09 –85.23%). Moisture was not affected by any of the studied variables.

Another interesting property that was analyzed, is solids separation or syneresis, that is influenced by process and solids incorporation. With a notable variety of values, it was slightly higher in whipped yogurt than in settled, attributed to the alteration of tridimensional structure or gel, that is a typical characteristic in settled yogurt, observing different trends in yogurt systems through storage time. This measure is also included, in Table 3. Yazici et al.[16] found that yogurt added with calcium salt (lactogluconate) exhibited higher syneresis than yogurt without this salt. In this study, the yogurt type resulted significative ($\alpha < 0.95$) on solids separation, at days 0, 7 and 14.

All these properties show a food item of high quality, in which the calcium salt addition did not affect importantly their properties but improve the nutritional value, more significant for women consumption.

Flow properties

These physical properties have been less characterized than other yogurt properties, there are few publications about it. These flow properties are very important for handling and processing of these liquid foods, in which the presence of several components, such fat, lactose, proteins, salts, and microorganisms in a lower degree, contribute to a non- Newtonian behavior.[2–4]

From viscometer data, shear rate and stress, the apparent viscosity was function of shear rate, showing a decreasing evolution that expresses the pseudoplastic nature of both yogurt types, with a flow behavior index lower than one. Even though a diversity of magnitudes for flow parameters, from rheograms, is clear that viscosity was higher (around three times) in settled yogurt than in whipped.

Fitting parameters from both rheological models (Eqs. 6 and 7) for kiwi yogurts are included in Table 4, showing very good fittings for both models: in which the yield stress showed clear differences, a range of 1.79 to 2.21 Pa for settled yogurt, in comparison with a range of 0.29 to 0.42 Pa for whipped one, that were computed from fitting to the second flow model or Herschel and Bulkley model. The viscosity of yogurts was relatively constant through storage, and it was influenced by calcium adding, in which the viscosity was lower than in yogurt control, with exception of yogurt with 150 mg.

Table 4 Rheological parameters for yogurt systems with kiwi flavor, at the beginning of the study

Yogurt system	Power Law Model			Herschel and Bulkley Model			
	n (dimensionless)	K (Pa sn)	R2	n (dimensionless)	K (Pa sn)	τo (Pa)	R2
300 mg MPh Settled/kiwi	0.55	67.64	0.98	0.55	66.15	1.83	0.98
250 mg MPh Settled/kiwi	0.54	62	0.99	0.55	60.46	1.79	0.99
150 mg DPh Settled/kiwi	0.52	77.57	0.99	0.52	75.95	1.89	0.99
125 mg DPh Settled/kiwi	0.4	162.6	0.99	0.4	160.5	2.21	0.99
300 mg MPh Whipped/kiwi	0.29	59.25	0.99	0.29	58.98	0.29	1
250 mg MPh Whipped/kiwi	0.42	29.56	1	0.42	29.15	0.42	1
150 mg DPh Whipped/kiwi	0.3	65.31	0.99	0.3	65.04	0.3	0.99
125 mg DPh Whipped/kiwi	0.39	58.82	1	0.4	58.41	0.4	1
Control settled-kiwi	0.46	138	0.99	0.46	136.1	2.14	0.99
Control whipped-kiwi	0.34	58.09	1	0.34	57.77	0.34	1

Sensory assessment

Finally, to know the human perception of prepared yogurts (control, 150, 250 and 300 mg/100mL), a sensory evaluation was completed, doing two types of evaluation a hedonic test and a triangular test.[11] For the first test, four sensory attributes were considered: appearance, flavor, texture, and overall acceptability.

Average evaluation, including the four attributes, showed a better qualification for both settled yogurt types and whipped kiwi; control yogurt was higher, followed by 250, 150 mg, and being 300 mg the last, as may be observed in table data. Average perception by consumers was decreasing with storage, although it was variable. Table 5 included the average evaluation of control yogurts for both flavors and other six yogurts, just to have a general idea; seven persons (not the same) completed the sensory evaluation each day.

Table 5 Average evaluation for control yogurt systems

Yogurt system	Day 0				Day 7				Day 14				Day 21			
	A	F	T	OA	A	F	T	OA	A	F	T	OA	A	F	T	OA
Control settled-kiwi	8.0	8.0	7.3	8.0	7.7	7.9	7.4	7.9	7.7	7.7	7.4	7.9	7.1	8.1	7.4	7.4
Control settled-p fruit	8.3	8.6	8.0	8.1	7.3	7.7	7.6	7.9	8.6	8.0	8.9	8.0	7.1	8.1	7.4	7.4
Control whipped-kiwi	7.6	6.9	7.0	7.1	7.7	7.1	7.1	7.1	7.3	6.9	6.7	7.3	7.9	7.3	6.9	7.1
Control whipped-p fruit	7.3	7.1	7.3	76	7.6	7.3	7.6	7.9	7.7	7.4	7.4	7.3	7.7	7.0	7.3	7.1
150 DPh whipped-kiwi	7.9	6.4	7.4	7.3	7.1	6.1	7.4	6.7	6.1	6.4	6.3	6.7	7.3	6.3	6.1	5.9
250 MPh whipped-kiwi	7.4	6.1	6.7	6.6	7.4	7.4	7.4	7.4	6.3	6.1	6.0	6.6	6.0	6.6	7.1	6.9
300 MPh whipped-kiwi	6.6	6.0	6.1	6.6	7.4	5.7	7.3	6.7	7.0	5.1	5.9	6.4	7.1	5.1	7.0	5.6
150 DPh settled-p fruit	7.1	7.1	7.3	7.1	7.1	6.6	6.6	6.6	7.0	6.9	7.1	6.4	7.3	6.1	5.4	6.0
250 MPh settled-p fruit	7.1	7.1	8.0	7.1	7.1	7.3	7.1	7.1	7.0	6.9	7.3	7.1	7.3	6.3	7.0	6.7
300 MPh settled-p fruit	6.1	5.6	7.0	6.6	7.1	6.3	7.6	6.9	7.0	6.1	7.1	6.3	7.3	6.3	7.0	6.7

These evaluations ranged from 5.6 to 8.9, showing amplitude and with most of the averages higher to six. Apparently, passion fruit was evaluated better than kiwi yogurts. Taking the results of statistical analysis, appearance did not have significant effect from storage time in days 0 and 7, being the contrary in days 14 and 21; flavor exhibited significant influence of yogurt type and calcium level on days 7 and 14, whereas on days 0 and 21, only had significance the calcium concentration. For texture, both factors, yogurt type and calcium level, influenced significantly on day 14, the other days did not. General acceptability, was only affected by yogurt type on day 0, and it was significantly influenced by calcium level on days 0, 7, 14 and 21.

Finally, a triangular test between the yogurts with 300 mg and control, let to know that added flavors favored the acceptation of both types due to masking of salty perception due to calcium salts. And even though, both yogurts (flavored and control) were perceived as different by five or more judges, they were significantly different from statistical viewpoint.

Conclusions

Yogurt as a nutritious food item, may be improved by the incorporation of calcium, then a complete and interesting work on yogurt with calcium was done, practical and usable data was generated through 21 days of study, in which different properties were measured. Yogurt with dibasic phosphate of calcium exhibited similar pH and acidity than control one, through elaboration process. Microorganisms grown was also similar the first three hours of elaboration, being different from the fourth hour, the number of colonies was higher in the enriched yogurt. Density and moisture were constant, without significant influence of yogurt type, neither salt type.

Acidity showed increasing during the storage time, in both settled yogurts (kiwi and passion fruit), as well as whipped of passion fruit. Syneresis resulted higher in whipped than in settled yogurt. Luminosity and b color parameter of yogurt were decreasing through storage time, in contrast to a color parameter. The net change of color was low for both yogurt types.

Both yogurt types exhibited a non-Newtonian response of shear thinning or pseudoplastic nature.

All yogurt samples were well qualified, being that of 300 mg/100mL the least accepted.

All samples of prepared yogurt had good stability through 21 days, without incorporation of a chemical preservative.

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None.

Conflicts of interest

The authors declare that there are no conflicts of interest.

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