

Development of a novel carob fortified yoghurt: Characterization of some Physicochemical and Organoleptic properties

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Abstract

The present study aim to investigate the effects of different concentrations (1, 1.5 and 2% ; w/v) of carob pulp flour and the whole carob pod powder on titratable acidity (% lactic acid), viscosity and organoleptic properties of yoghurt. For this purpose, eight different types of yoghurt were produced: 3 yoghurts with 1 to 2% of carob pulp flour, 3 yoghurts with the whole carob pod powder (including seeds and pulp) at 1 to 2% ratios and last 2 yoghurts with a mixture of 1.5 % of pulp carob flour and 1 to 1.5 % level of starch. The titratable acidity of yoghurts supplemented with 1 and 1.5 % ratios of carob pulp flour do not differ significantly from control. However, yoghurts fortified with the whole carob pod powder showed significantly ($p < 0.05$) higher titratable acidity than control. For viscosity, no significant differences ($p < 0.05$) were observed between control and yoghurts added with a mixture of 1.5% level of carob flour and 1 to 1.5% of starch levels. The highest sensory analysis scores were given by panelists to yoghurts enriched with 1.5 % of carob pulp flour and 1% of starch. Based on results of titratable acidity, viscosity and sensory properties of all yoghurts samples, we conclude that 1.5% level of carob pulp flour mixed with 1% level of starch is a suitable formulation for carob fortified yoghurt production.

Keywords: Carob pulp flour-the whole carob pod powder- carob fortified yoghurt- titratable acidity- viscosity- sensory analysis

1. Introduction

The dairy industry occupies a very important place among the agri-food industry in Morocco and over the world. Among the Fermented fresh dairy products, yougurt is the most consumed in many countries. The increase in the consumption of yoghurt is associated with its multiples health benefits and functional properties such as cholestertol reduction, protection against cancer, immunologic stimulation and improvement of gastro-intestinal functions^[1,2,3]. In addition, yoghurt constitutes an importante source of nutrients such as protein, fats and mineral especially for children and also for adults^[4].

Recently the development of a novel yoghurts formulation with natural health promoting substances has been attracting increase attention. The incorportaion of natural food additive, especially those containing fiber, into yoghurt production has been shown to improve its nutritional and sensory propeties for consumer acceptability^[5, 6, 7, 8]. Dietray fiber consumption has been suggested to control weight and serum cholesterol levels, reduce blood pressure and improve intestinal functions^[9, 10, 11]. Furthermore, some fibers have a prebiotic effect by their selective stimulation of one or more benficial bacteria in the colon^[12] and could also be used to improve the stability of probiotics microorganisms during food processing and storage^[13].

Carob pulp derived from *Ceratonia siliqua* pod, is classified the most rich food on insoluble dietary fiber, and polyphenols^[11, 14, 15]. Carob tree (*Ceratonia siliqua* L.) is a typical Mediterranean plant and Morocco is the fifth carob producer in the world after Spain, Italy, Portugal and Greece^[14, 16]. Because of their nutrients value and chemical

composition, seeds and pulp of carob fruit are used in Agroo-Food, pharmaceutical and cosmetics industries. Numerous human foods were prepared from the pulp such as sugar syrups or molasses, unroasted and roasted carob powder used as cocoa substitutes, or especially tannin rich preparations as anti-diarrheic products^[17, 18, 19]. In addition, seeds with high amount of galactomannans^[20] were used to extract carob gum. These constitute a valuable natural food thickeners, stabilizer and flavorant and is commonly added to a variety of product such as sweets and Ice cream^[21, 22]. Consumption of carob fiber has been proved to have a high anti-cardiovascular and antioxidant properties^[23] and to reduce serum cholesterol and serum triglycerides^[24]. Moreover, many studies have indicated that polyphenols may improve energy expenditure and fat oxidation in humans^[25]. Additionally, other sudies have shown that Carob fiber, consisting of polyphenols, strongly inhibits proliferation of both adenoma and adenocarcinoma cells of the human colon and reduce the viability of cervical cancer (HeLa) cells^[26, 27, 28].

Due to their biological importance and their strong association with health benefits, carob dietary fiber and polyphenols should be involved in the development of functional foods. Yoghurt as one of the most accepted and consumed product by children and adults, could stongly contribute to improve the consumption of Dietary fiber and polypphenols when prepared with carob pods.

The aim of the this study is to develop a novel yoghurt fortified with carob and to study the effect of different concentrations of Carob flour on the physicochemical and sensorial properties of yoghurt.

2. Materials and methods

2.1. Materials

Raw cow's milk was obtained from the dairy farms of Souss Massa region from Morocco. Milk used in each trial was subjected to various analyses to check the quality criteria, mainly titratable acidity (% lactic acid), density and fat measurements and antibiotic and alcohol tests

The dried carob pods collected from TATA region of the southern Morocco, were first cleaned and divided into two lots. The whole carob pod (seeds and pulp) of the first lot were powdered using an electric grinder and the obtained powder was then sieved and stored in glass jars protected from light, whereas, for the second lot, pods were first cracked open to remove all the seeds and the obtained pulp was grossly grinded and used as granular form.

2.2 Yoghurt processing

Yoghurts were prepared in the laboratory of Dairy industry named SILDA situated in Agadir-Morocco. Eight formulations of yoghurt were made (table 1). Three varieties, CP1, CP2 and CP3 were prepared with seedless carob pulp in granular form at 1%, 1.5% and 2% (w/v) levels, respectively. Three other yoghurt types, CSP1, CSP2 and CSP3, were made with the whole carob powder including seeds and pulp at the ratios of 1%, 1.5% and 2% (w/v) respectively. The last two other yoghurts, CPst1 and CPst2, were formulated from 1.5% level of carob pulp flour with 1 and 1.5% of starch, respectively.

For each formulation, the ingredients were added to the fresh raw milk (fat 3%, acidity 0.17%, viscosity 1.030) and mixed with 10% (w/v) of sugar and 3% (w/v) of skim milk powder. This mixture was pasteurized at 90°C for 5 min and left to cool at 45°C. Milk was then inoculated with 0.05% (w/v) of active starter culture and incubated at 44°C±1 until pH 4.7 was reached. After the coagulum was cooled, it was gently broken, placed into plastic cups and stored at 4°C. Yoghurt Formulations were carried out in triplicates. All yoghurt samples were subjected to acidity, viscosity and sensory tests after 10 h of storage.

2.3. physico-chemical analyses

2.3.1. Titratable acidity (% lactic acid)

The titratable acidity was estimated in all yoghurt samples by the method described by AOAC (1990) [29]. Approximately 10g of each yoghurt sample measured in a conical flask was diluted with 10ml of distilled water and titrated with the standardized 0.1N NaOH using phenolphthalein indicator, until a pink end point was observed. The titratable acidity was expressed as percentage of lactic acid according to the following formula:

$$\text{Acidity (\% lactic acid)} = 9 \text{ NV}_1/\text{V}_2$$

Where, V₁ = Volume (mL) of 0.1 N NaOH required for titration, N = Normality of NaOH used, V₂ = Volume (mL) of milk taken for test. (1 ml 0.1 N NaOH = 0.009 g lactic acid).

2.3.2. Apparent viscosity determination

The apparent viscosity of all yoghurt samples including control was recorded by using viscosimeter PCE-RVI. 2 at 10°C. Samples were stirred for 40 seconds before measurement. The viscosity values were expressed as millipascal-second [mPa·s].

2.4. Sensory analysis

The organoleptic properties of the carob yoghurt formulations, were evaluated by 10 trained panelists from the dairy industry SILDA, who are familiar with the characteristic qualities of produced yoghurts. The yoghurt samples were served to the panelists in plastic cups at 6°C and analyzed for appearance, colour, flavour, aroma, texture and overall acceptability. This analysis was performed using a nine-point hedonic scale [30] wherein a score of 1 represented «dislike extremely», score of 9 corresponded to «like extremely», and the middle score represented «neither like or dislike». Water was also provided to the panel members to cleanse their mouths before and after tasting each sample.

2.5. Statistical analyses

All analytical measurements were performed in triplicates and the results presented as a mean of the obtained values with the standard deviation. Data were treated by using one-way Analysis of Variance (ANOVA) to test the effects of different concentrations of carob pulp flour and the whole carob pod powder on yoghurt physico-chemicals and sensory properties. Tukey and Fisher LSD methods were employed to rank the groups when significant differences were found (P<0.05). Dunnett method was used to compare carob yoghurt formulations with control. Statistical analyses were made by using Minitab 18 Software.

3. Results and Discussion

3.1. Effect of carob addition on yoghurt titratable acidity (% Lactic acid)

The physicochemical properties of the yoghurt samples are given in table 1. Significant differences (p<0.05) were found between the different treatments for titratable acidity (% lactic acid) and viscosity values. In general, the highest titratable acidity values were observed in yoghurts fortified with carob flour. Yoghurts made with the whole carob pod powder had significant (p<0.05) higher values compared to yoghurt prepared from the pulp carob flour and yoghurt control. However, yoghurts supplemented with 1 and 1.5 % ratios of carob pulp flour (CP1, CP2, CPst1, CPst2) do not differ significantly from control.

Furthermore, titratable acidity of carob added yoghurt increased with increasing levels of carob. The highest values of lactic acid were recorded with yoghurts supplemented with 2% of carob flour. These findings are in agreement with those reported by Guler-Akin *et al.* (2016), who observed that acidity increased in the probiotic yoghurt ice cream supplied with 1% of carob extract and in those supplied with a mixture of 1% of carob extract and 1% of whey powder. This could be due to the carob components which promoted the growth and metabolic activity of lactic acid bacteria. Indeed, Guler-Akin *et al.* (2016) [31], reported that addition of carob extract and whey powder improved the survival of *L.acidophilus* and *Bifidobacterium BB-12* due to the possible prebiotic effects and antioxidant capacity of them. Furthermore, several other researches have proved the positive influence of plant components on growth and acidification ability of lactic acid bacteria [32, 33, 34]. Likewise, the increase of acid lactic value in yoghurt made with carob may also be explained by its content on fructose and glucose [21,35]. In fact, Amerinasab *et al.* (2015)

[33], demonstrated that an increase in date liquid sugar (DLS) content from 1 to 6% v/v led to a significant decrease in pH value ($p < 0.05$) of yoghurts samples and this was suggested to be due to the metabolic activity of starter culture which is promoted by providing the simple sugars of fructose and glucose in DLS. Similar results have been also reported by Cakmakci *et al.* (2014) [32] for set-type yoghurts supplied with carrot juice and sugar. However in the study of Moreira *et al.* (2017) [6], the addition of carob flour with refined sugar in the yoghurt preparation has led to an increase in the fermentation time. Also, a slow acid development was observed by Atasoy (2009) [36] in yoghurt enriched with carob juice concentrate and this was attributed to the slow growth of the starter due to the high content of sugar. These findings are in agreement with those of Moreira *et al.* (2017) [6] who found that the pH of yoghurt samples is gradually increased by increasing the DLS content from 6 to 9% v/v.

3.2. Effect of added carob on yoghurt viscosity

For the viscosity, results revealed a significant differences between the eight yoghurt formulations (Table 1). Indeed, the viscosity values varied from 1400 to 2000 mPa, s, and from 1860 and 2760 mPa, s for yoghurts supplemented with 1 to 2% levels of carob pulp flour and those prepared from 1 to 2% levels of the whole carob pod powder, respectively. For yoghurts made with a mixture of 1.5 % of carob and 1 to 1.5 % of starch, the viscosity fluctuated between 3400 and 3600

mPa.s. From these results we conclude that the viscosity increased as carob flour level increased. In fact, the viscosity values were higher for Yoghurts added with 2% of carob flour and those made with a mixture of 1.5% of carob flour and starch than those of the other carob added yoghurts. Similar finding was also reported by Guler-Akin *et al.* (2016) [31] for probiotic yoghurts ice cream containing 1% w/v of carob. This result was explained by the high water binding capacity of carob and its gum content which is used as valuable natural food thickeners and stabilizer [21, 22, 31]. In addition, others researches attributes the increase of yoghurt viscosity to the increase of fructose or glucose levels instead of sucrose and lactose concentrations [33, 37, 38]. According to Amerinasab *et al.* (2015) [33], an agglutination of fat globules and certain proteins was observed at low temperature when supplementing yoghurt with 1 to 6% of date liquid sugar.

Nevertheless, yoghurt added with a mixture of 1.5% of carob pulp flour and 1 to 1.5% of starch levels did not differ significantly from control sample containing 1.5% of starch and 0% of carob. In the other hand, carob added yoghurts without starch had significant ($p < 0.05$) lower viscosity value than control. However, according to the standard values of a Stirred yoghurt, viscosity should range between 2000 and 3000 mPa.s. Thus, the 2% level of carob flour could lead to avoid the use of starch in carob yoghurt preparation since their viscosity is within the range normally found in the stirred yoghurt.

Table 1 : Physico-chemical analyses of yoghurts supplemented with different levels of carob pod

Yogurt Code	level of Carob (%)	Starch level (%)	Acidity (%Lactic acid)	Viscosity (mPa, s)
Control	0,00	1,50	0,88 ±0,03 ^{c*}	3500±50,04 ^{a*}
CP1	1,00	0,00	0,9±0,03 ^{c*}	1400±23,09 ^d
CP2	1,50	0,00	0,91±0,03 ^{c*}	1510±34,64 ^d
CP3	2,00	0,00	1±0,01 ^b	2000±33 ^c
CSP1	1,00	0,00	1±0,02 ^b	1860±24,3 ^c
CSP2	1,50	0,00	1,2±0,02 ^a	2070±40,5 ^c
CSP3	2,00	0,00	1,205±0,023 ^a	2760±24,09 ^b
CPst1	1,50	1,00	0,85±0,04 ^{c*}	3400±98 ^{a*}
CPst2	1,50	1,50	0,85±0,02 ^{c*}	3600±40,5 ^{a*}

CP1, CP2 and CP3 = yoghurts produced with carob pulp flour ; CSP1, CSP2 and CSP3= yoghurts prepared with the whole carob pod powder ; CPst1 and CPst2 yoghurts prepared with 1.5 % of carob pulp flour and 1 to 1.5% of Starch ; C= yoghurt control without the addition of Carob.

^{abcd} Letters indicate significant differences between carob yoghurt formulations

* indicate No significant differences between carob yoghurt formulations and yoghurt control

3.3. Sensory analysis

Sensory characteristics such as appearance, colour, Flavour, Aroma, texture and overall acceptability, of the all yoghurt formulations were evaluated after 10 hours of storage. Effect of carob addition on the tested organoleptic properties is presented in table 2. It may be seen from the sensory scores that no significant difference ($p < 0.05$) was observed for appearance and colour between control and carob pulp fortified samples (CP1, CP2, CP3, CPst1 and CPst2), whereas a significant difference ($p < 0.05$) was revealed between control and yoghurts added with the whole carob powder. These one had the lower scores than control due to their brown colour. Similar trend was obtained by Guler-Akin *et al.* (2016) [31] who reported a decrease in the colour and appearance scores of carob extract supplemented ice creams because of its colour from orange to brown. For flavour and aroma, there were significant differences ($p < 0.05$) between control and carob supplemented yoghurts. Highest scores were given by the

panelists for the flavour of samples added with a mixture of carob pulp and starch followed by yoghurts enriched with carob pulp only. The flavour of the samples added with the whole carob pod scored less compared to other samples because of their bitter taste and sour acidity. The aroma scores for all carob added samples were higher compared to the control.

No significant differences ($p < 0.05$) were shown for texture among the control and all other fortified yoghurts, except for CSP1 sample made with 1% of the whole carob pod powder and samples CP1, CP2 added with 1 and 1.5% of the carob pulp flour, respectively. In fact, the texture of these samples scored less than other yoghurt samples. The overall acceptability scores of yoghurts supplied with carob pulp flour were higher than other yoghurt samples, but they did not differ significantly from control. Indeed, the highest scores was given to the CPst1 sample (8,8) followed by CP3 (8,3), control (8,1) and CPst2 (8) samples, in order. However, the other yoghurt formulations recorded lower

scores compared to the control. Our results are in agreement with those reported by Guler-Akin *et al.* (2016) [31] who showed that addition of carob extract and whey powder has improved the firmness, viscosity, mouth coating, taste, odour and general acceptability structure of probiotic yoghurts ice cream.

Table 2: Sensory properties of yoghurts supplied with different levels of carob pod

Sensory properties					
Yoghurt Code	Appearance and color	Flavour	Aroma	Texture	Over all acceptability
Control	8 ±0,82 ^{a*}	7,4 ±0,52 ^b	7,2±1,23 ^b	8,4±0,52 ^{a*}	8,1±0,74 ^{a*}
CP1	7,9 ±0,74 ^{a*}	8,4±0,52 ^a	8, ±0,63 ^a	5,9±1,20 ^b	6,4±0,70 ^b
CP2	8±0,67 ^{a*}	8,7±0,58 ^a	8,3±0,67 ^a	6±1,33 ^b	6,7±0,48 ^b
CP3	7,8±0,63 ^{a*}	8,5±0,85 ^a	8,2±0,63 ^a	8,2±0,42 ^{a*}	8,3±0,67 ^{a*}
CSP1	3,8±1,23 ^b	4±1,15 ^c	8,3±0,67 ^a	6,2±1,32 ^b	4,4±0,97 ^c
CSP2	4,4±1,84 ^b	3,5±1,27 ^c	8,2±0,63 ^a	8,2±0,63 ^{a*}	4,4±0,97 ^c
CSP3	4,3±1,77 ^b	3,5±1,18 ^c	8,3±0,67 ^a	8,3±0,48 ^{a*}	4,4±0,97 ^c
CPst1	8,4±0,52 ^{a*}	8,8±0,42 ^a	8,5±0,53 ^a	8,8±0,42 ^{a*}	8,8±0,42 ^{a*}
CPst2	8,2±0,79 ^{a*}	8,7±0,48 ^a	8,5±0,71 ^a	8,6±0,70 ^{a*}	8±0,47 ^{a*}

CP1, CP2 and CP3 = yoghurts produced with seedless carob pulp ; CSP1, CSP2 and CSP3= yoghurts prepared with the whole carob pod (seeds and pulp) ; CPst1 and CPst2 = yoghurts prepared with 1.5 level of carob pulp flour and 1 to 1.5% of Starch ; C= yoghurt control without the addition of Carob.

^{abc} Letters indicate significant differences between carob yoghurt formulations

* Indicate No significant differences between carob yoghurt formulations and yoghurt control

4. Conclusion

Carob pulp is considered as one of the functional food because of its high content on insoluble dietary fiber, polyphenols and natural vitamins. Consumption of carob fiber has been increasingly recognized for their antioxidant properties and their ability to reduce the serum cholesterol, serum triglycerides and risk of cardiovascular diseases. In order to improve the fiber consumption, fortification of yoghurt with carob was attempted. Based on results of physico-chemicals and sensory properties, the addition of 1.5 % level of carob with 1% level of starch to milk was selected as a best combination to enrich yoghurt. Indeed, yoghurt prepared from this formulation showed physicochemical properties comparable with commercial control sample and had the highest scores of sensory evaluation for appearance, colour, flavour, aroma, texture and overall acceptability. In conclusion, carob pulp could successfully be used in yoghurt production.

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6. Conflicts of interests

No Conflicts of interests are to declare

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