QUALITY EVALUATION AND SENSORIEL ATTRIBUTES OF STEAMED YOGHURT PREPARED BY CLARIFIED DATE LIQUID SUGAR

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ABSTRACT

The aim of this work was to obtain pure liquid sugars by clarification of date syrup and to evaluate the effect of the substitution of sucrose by liquid sugars on the qualities of the steamed yoghurt. One liter of distilled water at 80-85 °C was added to 200g of date during 2h, homogenized and filtered. The collected date syrup was clarified by bentonite which could result entirely clear liquid sugar. Production of liquid sugar with low levels of turbidity 45±0.4, viscosity 0.94±0.02 mPas, pectin 1.10±0.2%, dry matter 13.56±0.4%, proteins 0.17±0.3%, ash 0.16±0.03% and higher amounts of sugars 83.8±3% was successfully realized. This clarification was performed under same optimized conditions: concentration of bentonite 16.6 g.L⁻¹ and temperature 60 °C for 30 minutes. The inclusion of liquid sugar for manufacture of steamed yoghurts as substitute of sucrose found a good quality and high acceptability for its sensorial characteristics. These results suggest the potential use of liquid sugars for formulation of new food products.

Keywords: Bentonite, clarification, date, liquid sugar, yoghurt

INTRODUCTION

Date palm belonging to the Arecaceae family is the most arbocultural culture of the world (Sawaya et al., 1983). It has always played a principal role as food security crop of the people of arid and semiarid regions. The date fruit (Phoenix dactylifera L.) is very rich in nutritional substances such as: sugar, proteins, lipids, fibers, pectin, vitamins, and mineral salts (Ali-Farsi et al., 2007). Algeria is a date producer country. The varieties destined to the local consumption and to the export (Deglet-Nour) have morphological, microbiological and phycoschemical characteristics well-defined. Other varieties known as common dates (Himira) are low market value, negligible commercial interest, and limited economic importance (Ahmadnia and Sahari, 2008). The important quantities of the common dates do not meet the minimum quality characteristics for direct utilization. These varieties have been used as raw materials of fermentation industry or animal feed for many years. The non-use of this important by-product for human consumption constitutes a real economic loss. Date palm is very rich in sugars and sweetness; it can be used for substitution of granular sucrose for many food formulations. Different derivatives of dates (concentrate, syrup, powders, and liquid sugar) were used for substitution of sugar due to its richness in bioactive compounds and its economical value. Date was used in the formulation of yoghurt in the form of fiber (Hashim et al., 2009), syrup (Gad et al., 2010) and powder (Hariri et al., 2018). However, the texture and colour of novel products were modified. All forms of dates are in high viscous liquid phase with different physicochemical characteristics to granular sucrose and its addition in food provokes major problems for their packaging, transport, handing, piping and mixing with other ingredients. The addition of high viscous date into yoghurt is likely to change activity of starter culture. Date syrup contains different bioactive substances such as sugars, lipids, proteins, pectin and mineral salts (Alamazi, 2010). It can be obtained after elimination of the major colloidal substances. The obtained syrup is a sweet with a flavor similar to tough caramel (Elleuch et al., 2008). This syrup is used in formulation of confectionery, snacks and health beneficial foods. Liquid sugar can be prepared from date syrup after different steps of extraction, purification and elimination of colloidal substances such as protein, pectin, fiber and dyes (Ali-Farsi, 2003). Date liquid sugar, until now, have been not used in yoghurt formulation, this is the first work to use this natural sugar which is marked by an important nutritional value. This clarified natural product due to it purity can be used for the food formulations such as soft drink (Hariri et al., 2019) and yoghurt. Processes leading to form clarified by-products are delicate and largely depend on monitoring and controlling of various physical and chemical changes occurring during clarification process. Several methods of clarification of date liquid sugar were cited in the literature such as by utilization of conventional enzymes: pectinase and cellulase (Gamal A El Sharnoubiy, 2014), by removal of colored components using activated carbon ion-exchange resins (Nashti et al., 2012; Ahdno and Jafarizadeh-Malmiri, 2015), and by combined utilization of bentonite and gelatin (Jalali et al., 2014). Among the previously cited clarification methods, bentonite procures the high adsorption capacity due to important specific surface area. Efficiency of clarification by bentonite depends on various factors such as temperature, duration of clarification, concentration of this natural clay and pH of date palm syrup. The objective of this work was to investigate the optimization of the clarification procedure of liquid sugar from date palm syrup by utilization of bentonite and to determine the influence of substitution of sucrose by liquid sugar on the qualities of the steamed yoghurt.

MATERIAL AND METHODS

Primary materials

Chemical compounds

All solvents and chemicals substances were purchased form Sigma Aldrich.

Vegetable material

The common date fruits variety Himira (tamar stage of maturity) was collected from the region of Bechar (South-West of Algeria) in the month of September 2017. The selection of this low-grade date fruits was based on its low cost and its richness in sugar. The specie was identified by botanist at faculty of SNV, University of Mascara. The freshly edible date fruit (without seeds) was washed with distilled water, dried in darkness at room temperature and chopped into small pieces to increase the surface of diffusion.

Natural adsorbent

The bentonite used was obtained from the Magnhia deposit (West of Algeria) by national fat firm from the region of Sig (Mascara). This clay ready to use presented in the form of powders was used for the bleaching of edible oils. It is a silicate hydrates that contain quartz as the major impurity, and 85% of montmorillonite. This clay is a sodium bentonite with the following characteristics: pH 6.2, specific
area 80 mg m⁻², calcium (Ca²⁺) 30.60 meq.100g⁻¹, magnesium (Mg²⁺) 12.8 meq.100g⁻¹, sodium (Na⁺) 36.2 meq.100g⁻¹ and potassium (K⁺) 9.5 meq.100g⁻¹. This adsorbent presents strong capacity for ion-exchange and maximum capacity for adsorption of organic and inorganic matters.

### Extraction of date fruit syrup

One liter of distilled water at 80/85 °C was mixed with 200g of edible date fruit (cut into small pieces) during 2h, homogenized with a mixer Ultra-Turrax T25 (IKA, Germany). The collection of syrup was centrifuged for 10 min at 15000 rpm (Sigma labzentrifugen D-37620 Osterode am Harz, Germany) and left standing for 48h at room temperature to precipitate large quantities of insoluble substances and then filtered (Turhan et al., 2010). The final date syrup was filtered with Whatman filter paper (N°45).

### Clarification and physico-chemical characterization of liquid sugar

To obtain pure liquid sugar by clarification of date palm syrup, several methods were tested. The clarification by activated carbon gives small particles suspended in the date syrup, clarification by calcium oxide gives to the syrup a dark color, and clarification by bentonite found a satisfactory results. This method is influenced by several parameters such as: concentration of bentonite, duration of clarification, pH of the syrup, stirring speed and temperature. To optimize this method, four parameters were varied in a random way: concentration of bentonite from 12.5 to 50 g L⁻¹, duration of clarification (5 to 30 min), temperature (from 20 °C to 80 °C), and pH of date syrup (4.5, 5.2 and 9). The protocol OPAT (one factor at a time) was applied to optimize these four parameters.

#### Effect of the concentration of bentonite

The quantity of bentonite used was fixed at 5g and the volume of the date syrup was varied: 100 mL (concentration of 50 g L⁻¹), 200 mL (25 g L⁻¹), 250 mL (20 g L⁻¹), 300 mL (16.6 g L⁻¹), 350 mL (14.28 g L⁻¹) and 400 mL (12.5 g L⁻¹). The whole of the samples were carefully shaken and left at room temperature for 1, 3 and 6.5h. The choice of the optimal concentration of bentonite was based on the quantities of the residual ash and protein obtained after clarification.

#### Effect of the temperature and duration of clarification

The optimal concentration of bentonite was used and the temperature was varied at 20, 40, 60 and 80 °C. The residual ash and protein were determined after 5, 10, 15, 20, 25 and 30 min. The optical density was determined to verify the possibility of the Maillard reactions.

#### Effect of the pH

The optimal conditions of the clarification (concentration of the bentonite, duration and temperature of clarification) were applied. The quantity of the necessary absorbent was added to the date syrup at different pH (4, 5.2 and 9). The pH was regulated by addition of citric acid 0.1N to obtain pH 4 and by sodium hydroxide 0.1N to obtain pH 9. The bentonite and date syrup were agitated in a low speed and centrifuged for 7 minutes at 4500 rpm. The sweetened clarified syrup was concentrated in water bath at 105 °C until a viscous, clear, dark colored product was obtained. Physicochemical and biochemical characteristics of the date syrups (before clarification) and liquid sugars (after clarification) were determined. All samples were analyzed according to the AOAC method (AOAC, 2007). The pH was determined using a digital pH meter apparatus (Mettler Toledo. MP220) and turbidity by a turbidimeter Hana HI9370 model, America. The viscosity was evaluated on Giolitti Cantonii medium and Chapman agar after incubation at 37 °C for 24-48h for total coliforms and at 44 °C for faecal coliforms (Limas Tribst et al., 2009). Staphylococcus aureus was evaluated on Goliotti Cantoni medium and Chapman agar after incubation at 37 °C for 24-48h. Faecal streptococci were quantified in Rothe presumptive medium enriched by sodium azohydrate and Litsky confirmation medium added by sodium azohydrate and purple ethyl. Search sulfit-reducing Clostridium can be evaluated by counting the sporulated forms developed in media Meat Lister containing sodium sulphite and iron alum after incubation at 37 °C for 48h. Salmonella was evaluated in Salmonella agar medium incubated at 37 °C for 24-48h, after enrichment in Selenium-F Broth (SPB). The yeasts and moulds were quantified on potato dextrose agar medium (PDA) added by oxetaxylene after incubation at 25 °C for 5 days. To characterize the acceptability of the yoghurts stored after 21 days (Metin, 2006), all samples were analysed for sensory attributes by 10 panellists; using a point scale 5: good, 3: acceptable, 1: bad. The selected panelist was asked to fill in a questionnaire which included the following questions for the taste, texture and acidity. Panellists were informed of the type of product being tested and asked about their yoghurt consumption habits. Tap water was provided between samples to cleanse the palate.

### Statistical analysis

All experiments and analyses were done in triplicate. The results were statistically determined by analysis of variance (level of significance p<0.05) using SPSS Statistics software 8.1.

### RESULTS AND DISCUSSION

#### Clarification of date fruit syrup

### Effect of the concentration of bentonite

The results presented in figure 1, indicated significance (p<0.05) variable reduction of the proteins by the adsorbent according to the concentration used. The concentration 16.6 g L⁻¹ was able to reduce the proteins content from 4.1 ± 0.2% (initial content of date syrup) to 0.68 ± 0.1% after 3h of clarification (proteins adsorbed 83.40%). All concentrations of bentonite were able to reduce...
significantly the amount of the ash during the first hour of clarification from 0.38 ± 0.02% to very low values. A maximum amount of mineral salts was removed after 1h of clarification with the concentration of 12.5 g.L⁻¹. After 3h of treatment, an augmentation of the level of ash was observed until the initial value was exceeded for all samples. This increase can be explained by the nature of adsorbent which initially contains ionic groups. Increasing of it concentration allows the change in the pH and ionic strength of the solution which may have a negative effect on the adsorbed ash. From these results, the concentration chosen was 16.6 g.L⁻¹ which allows reducing the amounts of proteins and ash to acceptable values.

Effect of the pH

In order to maximize the elimination of the proteins and ash, and to minimize the optical density (preservation of the color and the sugar content of the clarified date syrups), we opted for the clarification for 30 min at 60 °C. The pH was adjusted at 4, 9, and without regulation (5.26). As shown in the table 2, the different steps of clarification significance (p≤0.05) remove the major substances except the sugars. The increase of the temperature has beneficial effect on the activity of the adsorbent and reduces the duration of clarification. Obtained results are similar to the values cited by Garg et al. (2004). The author’s affirmed the positive effect of the temperature on the adsorption of substances by bentonite. According to the same author’s, increasing the values of the temperature from 40 °C to 60 °C considerably improves the efficiency of the adsorption and led to reduction of viscosity.

Effect of the temperature and duration of clarification

From the results presented in table 1, it was noted that the heat treatment accelerates the activity of the adsorbent and reduces the duration of clarification. At low temperatures 20 °C and 40 °C and from the beginning of treatment, the clarified syrup displayed higher values of the optical density. The latter gradually decrease with the duration of treatment. This can be explained that at low temperatures, the solubility of suspended solids present in the syrups was low and therefore a higher optical density. The increase of the duration of treatment promotes the precipitation of these substances which reduces the optical density. The exposure of the syrups to higher temperatures 60 °C and 80 °C causes a lower optical density at the start of treatment and increases gradually with the time of clarification. This increase can be result to the Maillard reactions and therefore a darker coloration and a higher optical density of the syrups.

Table 1 Optical density (OD), residual proteins and ash after clarification at different times and temperatures of treatments

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Time (min)</th>
<th>OD at 420 (nm)</th>
<th>Proteins (%)</th>
<th>Ash (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>20</td>
<td>40</td>
<td>60</td>
<td>80</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>0.46±0.010</td>
<td>0.48±0.010</td>
<td>0.17±0.030</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>0.44±0.020</td>
<td>0.47±0.010</td>
<td>0.18±0.020</td>
</tr>
<tr>
<td>OD at 420</td>
<td>15</td>
<td>0.35±0.020</td>
<td>0.44±0.020</td>
<td>0.18±0.010</td>
</tr>
<tr>
<td>(nm)</td>
<td>20</td>
<td>0.29±0.010</td>
<td>0.34±0.020</td>
<td>0.18±0.010</td>
</tr>
<tr>
<td></td>
<td>25</td>
<td>0.27±0.010</td>
<td>0.20±0.020</td>
<td>0.20±0.020</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>0.11±0.020</td>
<td>0.17±0.020</td>
<td>0.21±0.010</td>
</tr>
</tbody>
</table>

**Koyuncu et al. (2007)** declared that the adsorption of substances that cause turbidity and colour in the apple juice is raised by bentonite with increasing temperature. The optimal conditions of clarification chosen (concentration of bentonite 16.6 g.L⁻¹, temperature 60 °C, time 30 min) were different to the conditions (concentration of bentonite 3 g.L⁻¹, pH 4.28, temperature 38 °C, time 82 min) cited by Jalali et al. (2014). This difference can be attributed to the nature and composition of bentonite used, variety of date treated, composition and method of extraction of date palm syrup. In our work, date syrup was obtained by hot extraction (concentration of date 200 g.L⁻¹, temperature 80-85 °C, time 2h). This method promote the maximum extraction of sugar but other macro and micro components such as proteins, pectin and mineral salts will also be extracted. For this, more severe conditions were used in terms of concentration of bentonite and temperature to eliminate the maximum of these undesirable substances and to
The date syrup has a very high turbidity (over 1000) due according to Junk and Pancost (1974) to the presence of large particles in colloidal suspension (total dietary fiber, pectin, polyphenolic compounds, coloured pigments, etc.) substituted during the hot extraction. After clarification, there was a sharp significance decrease in the level of turbidity to 45 ± 0.4 at pH 5.26. The decrease in pH of date syrup at value of 4 allows a strong significance decrease of the turbidity of the liquid sugar up to 22 ± 0.2. The protein content of the date syrup was 4.10 ± 0.20%, this low proportion was due to the poverty of dates in proteins. This data is higher to the results cited by several authors (Farahnaky et al., 2016; Gamal A El Sharnouby, 2014; El-Nagga and Abd El-Tawab, 2012) in date syrup obtained by different methods of extraction. After physical clarification, the liquid sugars presents total proteins respectively in the values of 0.07 ± 0.1 (at pH 4), 0.17 ± 0.3% (at pH 5.26) and 0.21 ± 0.2 (at pH 9). A part of the amino acids has waste groupings acidic or basic who are not involved in the peptide binding and which confers to the molecule the amphoteric character. The algebraic sum of the charges brought by a protein was its net charge which becomes zero at the pI (isoelectric pH). At the pH, the proteins will not be retained by the natural material clarifying used. At pH 4 (point very remote from the pI), the proteins were retained by the bentonite and were not eluted which explains the lower values of proteins and turbidity obtained. The decreasing values of proteins and turbidity of the date syrup by clarification leads to a clear liquid sugar. The level of pectin was 35.45 ± 0.3% in untreated date syrup. After clarification, the liquid sugar at pH 5.26 present very lower value 1.1 ± 0.2%. At pH 4 and 9, these values were remote of the pHpI which explains the lower quantity of pectin remaining after clarification. The obtained results indicated that the date syrup was rich in mineral salts 0.38 ± 0.02% but this value was lower to 2.23% cited by Farahnaky et al. (2016). After treatment, the liquid sugar displayed ash content in the value of 0.16 ± 0.03% (at pH 5.26). This significance decrease in the ash value is in line with the finding of El-Nagga and Abd El- Tawab (2012) and can be explained by the purification process and elimination of minerals, and insoluble substances. The filtration and defecation allow to eliminate a part of the mineral salts and the other part was inserted in the inter space leaf of the adsorbent. With the decreasing of pH, the ash content of clarified syrup has a non significant variation. This result is not in line with the results of Jalalit et al. (2014). The author’s showed that with decreasing pH, the ash content of syrup has a decreased. The results showed that the date syrup presents total sugar of 16.9 ± 0.3%. Similar values were obtained with other varieties of dates cited by Al-Farsi et al. (2007). After clarification and concentration, the results indicate a significance very high increase in the quantity of total sugars 84.2 ± 0.2% (at pH 4), 83.8 ± 0.3% (at pH 5.26), and 85.8 ± 0.1% (at pH 9). The clarification has allows an elimination of substances other than the sugars constituting the dry matter and the treatment of concentration eliminate the fractions of the free water. A similar result 81.88% obtained in liquid sugar was reported by Gamal A El Sharnouby (2014).

Inclusion of liquid sugar in steamed yoghurts

As shown in table 3, the TA of the CY evolves from 77 ± 0.2 °D in the first days to 102 ± 0.1 °D after 21 days of storage. The replacement of sucrose by liquid sugars causes a significance increase of the acidity during first day of the storage. The pH of the CY decreases significantly during time of the storage due to the acidification of the yoghurt by lactic acid bacteria. Yoghurts with liquid sugars present a slightly decreases of the value of pH during the first day. The pH evolves from 4.80 ± 0.1 for CY to 4.6 ± 0.2, 4.54 ± 0.1, and 4.49 ± 0.2 respectively for LSY20, LSY40 and LSY80. The decrease in the dry matter during time of storage was related to the utilization of these substances by the fermentations and microorganisms. After 21 days of storage, the dry matter content was 9 ± 0.2% for CY. The partial and total replacement of sucrose by liquid sugars significance decreases the level of the dry matter. During the storage period, the protein level of the CY decreases progressively from 4.4 ± 0.1% on the first day to 3.7 ± 0.1% after 21 days due to the intense multiplication of lactic fermentations and degradation leading to formation of soluble substances. The ash content of the CY decreases gradually during the storage period and reaches to value of 0.65 ± 0.2% after 21 days of storage. The sugars content present in the CY decreases from 130 g.1 to 68 g.1 after 21 days of storage. This decrease alters the nutritional quality of the steamed control yoghurt. The total and partial replacement of the sucrose by liquid sugar significance decrease the level of total sugar due to the lower volume of liquid sugar used. During the storage period, a very slight variation of the fat content was observed for all yoghurts prepared. The addition of the liquid sugar to the yoghurts provokes non significance change in the level of the fat because dates and liquid sugar are poor in the total fat. The TA of the CY increase during the storage period and contrary the pH decreases. Our result is similar to the pH cited by Hekmat and McMahon (1997).

This can be due according to Fanesar and Shinde (2011) to the consumption of sugar by lactic acid bacteria and formation of the lactic acid. The acidity can be increased by degradation of yoghurt proteins into free amino acids by proteolytic enzymes secreted by bacteria (Widyastuti and Febrisiantosa, 2014). This acidity increases during the substitution of sucrose by liquid sugar. This could be related to the large quantity of of reducing sugar in the liquid sugar which are more readily utilized by the lactic bacteria (Rubico et al., 2009) and are used by Yashaswini and Arunkumar (2016). The total protein content obtained was higher than 3.7 g.100g-1 and 3.2 g.100g-1 respectively reported by Rubico et al. (1987) and Buttirss (1997). Our results are in line with the results of Gündoğdu et al. (2009) who reported that the proteins content of yoghurts changed between 4.13 and 4.19% and decreased during the storage time. The replacement of sucrose by liquid sugar causes no significance changes in the level of the proteins content. According to Yashaswini and Arunkumar (2016) the addition of date syrup showed no effect on the level of fat and proteins of the tested yoghurt. Gad et al. (2010) affirmed that the inclusion of 10% of date palm syrup to the yogurt causes a decrease of the values of proteins, acidity and moisture. The level of ash obtained was higher to 0.27 g 100g-1 cited by Isanga and Zhang (2009). Results of microbiological evaluation showed absence of the Staphylococcus aureus, faecal streptococci, total and fecal coliforms, and Salmonella in all tested yoghurts and throughout the time of storage. This good microbiological quality can be explained by the effectiveness of heat treatments, the respect of hygienic conditions, and the acidifying activity of the lactic acid bacteria which inhibits the development of these germ. The yeast and moulds appear in the first week of storage (2 yeasts.mL-1), then this microorganisms increases to 4 yeasts.mL-1 during the last week of the storage for CY. Yoghurts with liquid sugar were marked by the presence of the similar number of yeasts and moulds (3 in the first week and 5 after 21 day of storage). The results of microbiological characterization for all steamed yoghurts prepared are in accordance with the results of Olmedo et al. (2013).
The sensory characteristics were carried out on the acidity, taste and texture of the yoghurts. As shown in figure 2, the CY was first appreciated by the panelist and has the significance good acidity (averages notation of 5.0 ± 0.2), followed by LSY20, then LSY40 and LSY80 respectively with average notations of 4.7 ± 0.1, 4.5 ± 0.2 and 4.4 ± 0.2. Further increase of liquid sugar decreases significantly the acidity due to the enhanced characteristics sweet flavour of liquid sugar which will mask the acidity of the yoghurt. The members of the jury appointed the yoghurts treated by high volumes of the liquid sugar (LSY40 and LSY80) as the yoghurt with the significance best texture and taste. The results of sensory evaluation of the yoghurts are in correlating with the work of Yashaswini and Arunkumar (2016). According to Gad et al. (2010) and Yashaswini and Arunkumar (2016), the addition of date syrup up to 10% level increases the sensory attributes with good biochemical, microbiological and sensory characteristics of the steamed yoghurts (LSY40 and LSY80) as the yoghurt further increase of liquid sugar.

CONCLUSION

Elaboration of liquid sugars from date syrup can extend the utilization of date by-products as substitutes for sucrose as a sweetener in different foods. Date liquid sugar was successfully manufactured by the use of bentonite, clarification procedures, and concentration of date syrup. Clarification of date liquid sugar with low quantities of turbidity, proteins, pectin, ash and higher levels of sugar was successfully realized under same conditions (concentration of bentonite 16.6 g L⁻¹, temperature 60 °C, time 30 min, pH of the date syrup close to 4). The use of liquid sugar for manufacture of steamed yoghurts as substitute of sucrose found a good quality with good biochemical, microbiological and sensory characteristics. These results suggest the potential use of liquid sugars for formulation of new food products.

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Conflicts of interest: The authors declare no conflict of interest.

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