Physico-Chemical and Functional Properties of Functional Yoghurt Made With Different Types of Whey Protein Concentrates (WPC).

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Abstract

Functional yoghurt was made using different types of whey protein concentrate (WPC). The chemical, physical, microbiological and sensory quality of the produced functional yoghurt were studied. The obtained results revealed that slight differences were observed for acidity, pH, TS, protein, fat and TVFA either for storage periods or between the treatments. Also, the obtained results revealed that T8 (functional yoghurt made with particulated whey protein at pH 5) recorded higher content of TVFA and acetaldehyde followed by T10 (functional yoghurt made with commercial whey protein concentrate powder) either when fresh or during the interval storage periods. The previous results reflected the good properties of physical characteristic of the produced functional yoghurt through increasing the viscosity, WHC and reduce the syneresis which affected by the amount of added wpc. On the other hand, the sensory evaluation results indicated that all the produced functional yoghurt were good in their characteristics. The highest scores were recorded after 7 days of storage at 5°C. Also, T9 and T10 which contain 2 and 3% wpc recorded 93.33 and 94.33, respectively.

Key words: functional yoghurt, WPC, TVFA, phyico-chemical properties.

Introduction

Yoghurt is one of the best known fermented dairy products which contain probiotic. Yoghurt is defined by the Codex Alimentarius 2003 as a coagulated milk product that results from the fermentation milk by lactic acid bacteria i.e. Streptococcus thermophilus and Lactobacillus delbrueckii subsp. bulgaricus.

Yoghurt products have achieved considerable economic importance worldwide owing to their high nutritional image and it has many health benefits such as improving lactose intolerance, anticholesterolemic impacts and reducing risk cancers and other benefits related to probiotics bacteria…..etc. (Laiho et al., 2017). The allegedly hypercholesterolemic effect of milk fat and the desire to ensure overall good health have led consumers to demand reduced fat milk dairy products, mainly including yoghurt, to reduce the risk of coronary heart disease (Sandoval, et al., 2004).

Demand for low milk fat / fat free milk with similar sensory properties with conventional full-fat products. Additional of functional ingredients such as whey protein concentrate (WPC) may improve overall quality of low-fat yoghurt. It is well known ability of (WPC) to support formation of whey protein aggregates which highly improve physical properties of yoghurt (Mikal et al., 2012).

Whey is considered a valuable product because of its soluble proteins and its high levels of amino acid, B vitamins, lactose and salts. (Barbosa et al., 2010). High nutritional quality, potent biological activity and unique functional properties are the foremost attributes of whey proteins (wps) that help sustain interest in their utilization, not only in the food industry but also in allied areas such as the pharmaceutical and bio-medical field (Battacharjee et al., 2006).

So, the aim of this study, to use whey protein concentrate (WPC) for manufacture of functional yoghurt for their nutritional and functional properties and to examine the effect of adding different concentration of (WPC) as a fat replacers on the chemical, physical and functional properties of the produced yoghurt.

Materials and methods

Materials

Bacterial strains and ingredients:-

a- Whey: Cheddar cheese whey used in this study was obtained from Arab Dairy Co (kaha, kalubiya Governorate, Egypt) and was used for making WPC.

b- Milk: Fresh mixed milk (cows and buffalos) were obtained from the heard of Faculty of Agriculture, Moshtohor, Benha University, Egypt.

c- Whey protein concentrate powder was obtained from Davisco Foods International, Inc, 11000Weast 78 the Street, Suite 210 Eden Prairie, Minnesota and purched from local markt.

d- Yoghurt starter cultures consisting of Lactobacillus delbrueckii subsp bulgaricus and Streptococcus thermophilus were obtained from Chr. Hansen’s Laboratories, Copenhagen, Denmark, and purched from local market.

Preparation of whey protein concentrate by Ultrafiltration:

Whey protein concentrate (WPC) was prepared using Ultrafiltration technique as follows: pH of whey adjusted to (6.5), the whey...
concentrated using cross-flow zirconium-titania ceramic membranes (50 kDa cut-off and 0.020 m² effective membrane areas). The inlet and outlet pressures were adjusted and controlled to 3.0 bar, and 5.0 bar, respectively. The temperature was maintained at ~40°C. The concentration process stopped when the whey reached to ~ 14 - 15 % total solids.

Preparation of whey protein / carrageenan particulate:

The method of Shenana et al., (2007). Was used for preparation of whey protein particulate as follows: (a) sodium carrageenan was added to the whey retentate at a rate of 0.1 g 100g (w/w). The pH was adjusted to pH 5 as a treatment and pH 3 using 6N HCl. (b) the whey retentate containing the carrageenan was heated at 85°C for 30 min, and then homogenized at 60°C using 2 stages laboratory homogenizer (Rannie, Copenhagen) at 20 and 5 MPa for the 1st and 2nd stage, respectively and (c) the homogenized retentate/ carrageenan mixture was then centrifuged at 5000 G for 10 min. The precipitate was considered as particulate whey protein/carrageenan concentrate (PWPC).

Manufacture of functional yoghurt:

Fresh mixed milk was standardized to fat content ~ 3% (full) and ~ 1.5 % fat (low), and used for manufacture of functional yoghurt according to the method described by Tamime, (1978). Treatments were prepared as follows:-

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3%fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat).
T1: Low-fat mixed milk cows and buffalos (1:1) ~1.5%fat + 2% with whey protein.
T2: Low-fat mixed milk cows and buffalos (1:1) ~1.5%fat + 3% with whey protein
T3: Low-fat mixed milk cows and buffalos (1:1) ~1.5%fat + 2% with whey protein concentrate.
T4: Low-fat mixed milk cows and buffalos (1:1) ~1.5%fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% with particulated whey protein concentrate at pH3.
T6: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% with particulated whey protein concentrate at pH3.
T7: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% with particulated whey protein concentrate at pH5.
T8: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% with particulated whey protein concentrate at pH5.
T9: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% commercial whey protein concentrate powder.
T10: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% commercial whey protein concentrate powder.

Analytical methods:

Total solids, ash, fat and total protein were determined according to the AOAC (2012). Titratable acidity was determined according to the methodology described by BSI (2010). pH values were measured using a digital laboratory pH meter (model HANNA pH 213 instruments) with combined glass electrode. Total volatile fatty acids (TVFA) contents were determined by the direct distillation method as described by Kosikowski, (1984). Acetaldehyde content was determined according to the method described by Lees and Jago (1969).

Microbiological examinations:

Total viable counts (TVC) of the produced functional yoghurt were determined as described by IDF (1991). Yeasts and moulds counts were done as described by IDF (1990). Coliform groups were detected according the methods of APHA (1992).

Rheological analysis:

Both syneresis and water holding capacity (WHC) were estimated according to modified method of Keogh and O’Kennedy (1998). The apparent viscosity was measured according to Petersen et al., (2000). The viscosity of the produced functional yoghurt was measured, after stirring the product for 60s, using a Brookfield viscometer model RVDVE (Brookfield Engineering Laboratories Inc., Middleboro, MA) at 10°C. Samples were tested using spindle no. 4 and data were taken as duplicate at a spindle rotation of 12 rpm.

Sensory evaluation:

Sensory properties of the produced functional yoghurt with wpc were done according to the scheme of (Tamime and Robinson, 1999) when fresh and during the storage periods up to 21 days by 10 staff of dairy sci. Dept. faculty of agri; Benha univ., Egypt.

Results and discussion

Coagulation time:-

The effect of adding different percentage and different types of wpc on the coagulation time of functional yoghurt are presented in table (1). The obtained results revealed a slight decrease of coagulation time of yoghurt by increasing the wpc addition, this may be due to the presence of whey protein which increase the activity of starter culture bacteria. The results are in accordance with El-Alfy et al., (2018).
Table 1. Coagulation time of functional yoghurt made with different percentage of WPC.

<table>
<thead>
<tr>
<th>Replicates</th>
<th>Treatments</th>
<th>Coagulation time / h</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C1</td>
<td>C2</td>
</tr>
<tr>
<td>1st replicate</td>
<td>3.36</td>
<td>3.27</td>
</tr>
<tr>
<td>2nd replicate</td>
<td>2.49</td>
<td>2.42</td>
</tr>
<tr>
<td>3rd replicate</td>
<td>3.26</td>
<td>3.20</td>
</tr>
</tbody>
</table>

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3% fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat).
T1: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein.
T2: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein.
T3: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein concentrate.
T4: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at pH 3.
T6: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at pH 3.
T7: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at pH 5.
T8: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at pH 5.
T9: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% commercial whey protein concentrate powder.
T10: Low fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% commercial whey protein concentrate powder.

Chemical composition of functional yoghurt:
Acidity and pH values:
Table (2) shows the average levels of acidity and pH of functional yoghurt with different percentage of WPC compared with the control low-fat and full-fat content. The mean values of acidity of the T5 and T10 were slightly higher than those obtained for the control and other treatments when fresh and along the storage periods up to 21 days at 5°C. This may be attributed to the acidity of WPC used in the both two treatments. Titratable acidity of all treatments increased gradually all over the storage periods up to 21 days due to the activity of the starter culture bacteria. These results are in agreement with those of Zedan et al., (2001).

Table 2. Acidity% and pH values of functional yoghurt made with different percentage of WPC when fresh and during the storage periods up to 21 days at ~ 5°C.

<table>
<thead>
<tr>
<th>Storage period days</th>
<th>Treatments</th>
<th>Acidity%</th>
<th>pH value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C1</td>
<td>C2</td>
<td>T1</td>
</tr>
<tr>
<td>Fresh</td>
<td>0.71</td>
<td>0.71</td>
<td>0.65</td>
</tr>
<tr>
<td>7 days</td>
<td>0.76</td>
<td>0.75</td>
<td>0.73</td>
</tr>
<tr>
<td>14 days</td>
<td>0.77</td>
<td>0.78</td>
<td>0.75</td>
</tr>
<tr>
<td>21 days</td>
<td>0.76</td>
<td>0.79</td>
<td>0.75</td>
</tr>
<tr>
<td>7 days</td>
<td>4.29</td>
<td>4.31</td>
<td>4.32</td>
</tr>
<tr>
<td>14 days</td>
<td>4.18</td>
<td>4.17</td>
<td>4.27</td>
</tr>
<tr>
<td>21 days</td>
<td>4.11</td>
<td>4.13</td>
<td>4.18</td>
</tr>
</tbody>
</table>

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3% fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat).
T1: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein.
T2: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein.
T3: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein concentrate.
T4: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at pH 3.
T6: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at pH 3.
T7: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at pH 5.
T8: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at pH 5.
T9: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% commercial whey protein concentrate powder.
T10: Low fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% commercial whey protein concentrate powder.
The pH values took an opposite trends to that of acidity, the corresponding pH values of both T3 and T5 were slightly lower than those of control and other treatments the pH values of all treatments decreased during the storage period due to the increase of starter activity and acidity, the present results are in agreement with those given by Nahed EL-Wahsh (2013).

**Gross chemical composition:**

Table (3) shows the average gross chemical composition of functional yoghurt made with different types of wpc when fresh and during storage periods up to 21 days. In general the total solid contents of low-fat yoghurt were lower than that of full-fat yoghurt, due to the high fat content of the control full-fat yoghurt.

<table>
<thead>
<tr>
<th>Storage period (day's)</th>
<th>C1</th>
<th>C2</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh</td>
<td>13.31</td>
<td>12.25</td>
<td>11.90</td>
<td>12.02</td>
<td>11.99</td>
<td>12.18</td>
<td>11.93</td>
<td>12.16</td>
<td>11.81</td>
<td>11.90</td>
<td>11.74</td>
<td>11.84</td>
</tr>
<tr>
<td>21 days</td>
<td>13.52</td>
<td>12.47</td>
<td>12.27</td>
<td>12.18</td>
<td>12.50</td>
<td>12.51</td>
<td>12.21</td>
<td>12.44</td>
<td>12.21</td>
<td>12.1</td>
<td>12.16</td>
<td>12.43</td>
</tr>
</tbody>
</table>

| Protein               | Fresh | 3.99 | 3.94 | 3.73 | 3.93 | 3.8 | 3.97 | 3.81 | 3.94 | 3.86 | 3.92 | 3.96 | 4.04 |
|                       | 7 days | 4.03 | 4.01 | 3.80 | 3.98 | 3.91 | 4.03 | 3.92 | 4.02 | 3.9 | 4.01 | 4.02 | 4.08 |
|                       | 14 days | 4.08 | 4.04 | 3.93 | 4.01 | 4.05 | 4.21 | 3.94 | 4.13 | 3.95 | 4.09 | 4.05 | 4.13 |
|                       | 21 days | 4.15 | 4.11 | 3.98 | 4.05 | 4.10 | 4.25 | 4.14 | 4.26 | 4.11 | 4.28 | 4.16 | 4.25 |

| Ash                   | Fresh | 0.75 | 0.76 | 0.71 | 0.74 | 0.75 | 0.77 | 0.72 | 0.74 | 0.71 | 0.72 | 0.68 | 0.70 |
|                       | 7 days | 0.79 | 0.78 | 0.78 | 0.79 | 0.79 | 0.82 | 0.75 | 0.75 | 0.73 | 0.76 | 0.73 | 0.74 |
|                       | 14 days | 0.85 | 0.84 | 0.8 | 0.83 | 0.83 | 0.84 | 0.82 | 0.83 | 0.79 | 0.81 | 0.76 | 0.77 |
|                       | 21 days | 0.86 | 0.86 | 0.83 | 0.87 | 0.86 | 0.84 | 0.82 | 0.84 | 0.84 | 0.78 | 0.79 |

| Fat                   | Fresh | 3.00 | 1.50 | 1.50 | 1.50 | 1.5 | 1.49 | 1.48 | 1.50 | 1.52 | 1.53 | 1.60 | 1.58 |
|                       | 7 days | 3.02 | 1.51 | 1.52 | 1.50 | 1.5 | 1.49 | 1.49 | 1.50 | 1.56 | 1.53 | 1.68 | 1.69 |
|                       | 14 days | 3.05 | 1.57 | 1.53 | 1.53 | 1.50 | 1.52 | 1.51 | 1.55 | 1.62 | 1.60 | 1.69 | 1.69 |
|                       | 21 days | 3.02 | 1.60 | 1.52 | 1.57 | 1.56 | 1.54 | 1.53 | 1.58 | 1.62 | 1.60 | 1.68 | 1.73 |

| TVFA                  | Fresh | 7.67 | 6.33 | 6.83 | 7.33 | 7.17 | 7.83 | 8.00 | 8.67 | 8.17 | 9.00 | 8.50 | 8.83 |

| Acetaldhyde (µg/100g) | Fresh | 17.66 | 17.65 | 17.71 | 17.73 | 17.69 | 17.70 | 17.75 | 17.78 | 17.76 | 17.79 | 17.62 | 17.69 |
|                       | 7 days | 17.83 | 17.71 | 17.74 | 17.79 | 17.74 | 17.78 | 17.83 | 17.79 | 18.75 | 17.93 | 17.64 | 17.70 |
|                       | 14 days | 17.57 | 17.59 | 17.71 | 17.60 | 17.59 | 17.58 | 17.62 | 17.73 | 17.71 | 17.58 | 17.63 |
|                       | 21 days | 17.56 | 17.55 | 17.54 | 17.57 | 17.55 | 17.56 | 17.58 | 17.62 | 17.59 | 17.63 | 17.5 | 17.53 |

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3%fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat).
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T4: Low-fat mixed milk cows and buffalos (1:1) ~1.5%fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% with particulated whey protein concentrate at pH3.
T6: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% with particulated whey protein concentrate at pH3.
T7: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% with particulated whey protein concentrate at pH5.
T8: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% with particulated whey protein concentrate at pH5.
T9: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 2% commercial whey protein concentrate powder.
T10: Low-fat mixed milk cows and buffalos (1:1) 1.5%fat + 3% commercial whey protein concentrate powder.
The total solids of functional yoghurt containing \textit{wpc} were lower than the corresponding values of the control either when fresh or along the storage periods up to 21 days this could be attributed to the low solids content of the \textit{wpc} and high water holding capacity of \textit{wpc}. On the other hand, the total solid of the control and different treatments increased gradually all over the storage periods up to 21 days and this could be due to the limited water losses during storage periods Shenana \textit{et al.}, (2007).

The protein content of functional yoghurt was slightly increased in a proportional rate with the addition of \textit{wpc}. This can be attributed to the high protein content of \textit{wpc} Shenana \textit{et al.}, (2007).

The fat and ash contents of the functional yoghurt was nearly not affected by the addition of \textit{wpc} as its almost free of fat Mehanna and Gone (1988).

The protein, fat and ash contents of control and different treatments of functional yoghurt were slightly increased all over the storage periods up to 21 days corresponding to the increase of total solids of different treatments Shenana \textit{et al.}, (2007).

The total volatile fatty acids contents of control and functional yoghurt from different treatments were gradually increased all over the storage period up to 21 days. This can be related to the growth and activity of the lactic acid starter culture similar trend was reported by Shenana \textit{et al} (2007) and El-Dahma (2018). The produced functional yoghurt with \textit{wpc} (T4) recorded higher (TVFA) content comparing with the control and the other different treatments.

The acetaldehyde content of controls and functional yoghurt of different treatments were gradually decreased by prolonging the storage periods up to 21 days at ~5°C. This decreased attributed to acetaldehyde is transitory component, where it begins in decrease after the fermentation is complete in the first 5 hour of storage. (Hamad \textit{et al.}, 2016).

\textbf{Rheological properties:}

Table (4) shows the physical properties of functional yoghurt with different types of \textit{wpc} when fresh and during storage at ~5°C.

<table>
<thead>
<tr>
<th>Storage periods (day’s)</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>C1</strong></td>
<td>16523</td>
<td>16133</td>
<td>16050</td>
<td>16540</td>
<td>16453</td>
<td>16613</td>
<td>16556</td>
<td>16456</td>
<td>16520</td>
<td>16570</td>
</tr>
<tr>
<td><strong>C2</strong></td>
<td>16453</td>
<td>16400</td>
<td>16426</td>
<td>16416</td>
<td>16560</td>
<td>16580</td>
<td>16556</td>
<td>16650</td>
<td>16596</td>
<td>16630</td>
</tr>
<tr>
<td><strong>T1</strong></td>
<td>16856</td>
<td>16766</td>
<td>16400</td>
<td>16610</td>
<td>16533</td>
<td>16583</td>
<td>16343</td>
<td>16396</td>
<td>16586</td>
<td>16574</td>
</tr>
<tr>
<td><strong>T2</strong></td>
<td>16866</td>
<td>16600</td>
<td>16563</td>
<td>16623</td>
<td>16600</td>
<td>16660</td>
<td>16473</td>
<td>16566</td>
<td>16453</td>
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<table>
<thead>
<tr>
<th>Storage periods (day’s)</th>
<th>C1</th>
<th>C2</th>
<th>C3</th>
<th>C4</th>
<th>C5</th>
<th>C6</th>
<th>C7</th>
<th>C8</th>
<th>C9</th>
<th>C10</th>
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</thead>
<tbody>
<tr>
<td><strong>Fresh</strong></td>
<td>31.81</td>
<td>31.41</td>
<td>25.82</td>
<td>29.64</td>
<td>30.01</td>
<td>27.95</td>
<td>24.82</td>
<td>27.85</td>
<td>26.71</td>
<td>24.05</td>
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<tr>
<td><strong>7 days</strong></td>
<td>33.89</td>
<td>33.89</td>
<td>28.51</td>
<td>31.01</td>
<td>29.77</td>
<td>30.16</td>
<td>27.92</td>
<td>25.86</td>
<td>28.32</td>
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<td><strong>14 days</strong></td>
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<td>34.30</td>
<td>29.43</td>
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<td>30.91</td>
<td>30.40</td>
<td>31.03</td>
<td>32.77</td>
<td>28.90</td>
</tr>
<tr>
<td><strong>21 days</strong></td>
<td>36.96</td>
<td>35.82</td>
<td>30.88</td>
<td>32.17</td>
<td>32.78</td>
<td>34.36</td>
<td>31.78</td>
<td>30.80</td>
<td>33.40</td>
<td>32.50</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Storage periods (day’s)</th>
<th>Water holding capacity (WHC %)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Fresh</strong></td>
<td>48.85</td>
</tr>
<tr>
<td><strong>7 days</strong></td>
<td>47.5</td>
</tr>
<tr>
<td><strong>14 days</strong></td>
<td>46.71</td>
</tr>
<tr>
<td><strong>21 days</strong></td>
<td>44.08</td>
</tr>
</tbody>
</table>

\textbf{Table 4.} Physical properties of functional yoghurt made with different treatments of WPC when fresh and during storage at ~5°C.

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3% fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat).
T1: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein.
T2: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein.
T3: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 2% with whey protein concentrate.
T4: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5% fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at PH3.
T6: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at PH3.
T7: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% with particulated whey protein concentrate at PH5.
T8: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% with particulated whey protein concentrate at PH5.
T9: Low-fat mixed milk cows and buffalos (1:1) 1.5% fat + 2% commercial whey protein concentrate powder.
T10: Low fat mixed milk cows and buffalos (1:1) 1.5% fat + 3% commercial whey protein concentrate powder.
On the other hand, the results recorded a slower syneresis and higher water holding capacity values comparing with the control. The decrease of syneresis was proportional to the increase of whey protein concentrates which increases water holding capacity due to the increase of protein matrix. The present results agree with that obtained by (Das and Seth, 2017) and El-Alfy et al. (2018).

Microbiological properties:

**Total viable bacterial counts (TVC)**

Table (5) shows that the total viable bacterial counts (TVC) of functional yoghurt from different types of wpc. TVC were not affected by both two levels of wpc, added to different treatments for fresh functional yoghurt. The corresponding number of TVC were slightly increased during storage for all the treatments. The same trend of results was also observed during the storage periods of functional yoghurt up to 21 days but the high increase of viable counts were observed within the first 7 days of storage, then followed by slightly decrease again among different treatments up to the end of storage and this may be attributed to the development of acidity by the starter cultures which affect the activity of bacteria. Nahed El-Wahsh (2013).

The yeasts and moulds were not detected in all fresh yoghurt either the control or functional and within the first 7 days of storage very low counts less than 10 cfu of yeasts and moulds were detected after 14 and 21 days of storage, respectively. Similar results were obtained Nahed El-Wahsh (2013), Coliform groups were not detected of all functional yoghurt when fresh and all over the storage periods. This indicates the good sanitary and hygienic conditions during making the products Shenana et al., (2007).

<table>
<thead>
<tr>
<th>Storage periods (day’s)</th>
<th>C1</th>
<th>C2</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh</td>
<td>7.00</td>
<td>7.14</td>
<td>7.05</td>
<td>6.99</td>
<td>6.88</td>
<td>6.73</td>
<td>6.87</td>
<td>6.91</td>
<td>7.05</td>
<td>7.04</td>
<td>6.73</td>
<td>7.12</td>
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<tr>
<td>7 days</td>
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<td>7.4</td>
<td>7.34</td>
<td>7.24</td>
<td>7.2</td>
<td>7.42</td>
<td>7.37</td>
<td>7.07</td>
<td>7.16</td>
<td>7.42</td>
<td>7.23</td>
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<tr>
<td>14 days</td>
<td>6.88</td>
<td>7.37</td>
<td>6.69</td>
<td>7.23</td>
<td>7.12</td>
<td>7.13</td>
<td>7.2</td>
<td>7.03</td>
<td>7.09</td>
<td>7.2</td>
<td>7.05</td>
<td>7.3</td>
</tr>
</tbody>
</table>

C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3%fat).
C2: Control (low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat).
T1: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 2% with whey protein.
T2: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 3% with whey protein.
T3: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 2% with whey protein concentrate.
T4: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 3% with whey protein concentrate.
T5: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 2% with particulated whey protein concentrate at pH3.
T6: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 3% with particulated whey protein concentrate at pH3.
T7: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 2% with particulated whey protein concentrate at pH5.
T8: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 3% with particulated whey protein concentrate at pH5.
T9: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 2% commercial whey protein concentrate powder.
T10: Low-fat mixed milk cows and buffalos (1:1) ~ 1.5%fat + 3% commercial whey protein concentrate powder.

Sensory evaluation:

All prepared functional yoghurt treatments were evaluated when fresh and during interval storage (~ 5°C) up to 21 days for the different organoleptic properties including flavour, body & texture and appearance. The panelist score are presented in Table (6). In general it can be concluded that the different treatments recorded slightly higher score than the control, also the functional yoghurt containing whey protein concentrate (T5) ranked the highest score points when fresh and during the first 7 days of storage. After 14 and 21 days of storage the organoleptic scores of all samples were gradually decreased, and the lowest values were observed at 21 days of storage this could be attributed to the increase of acid development as recorded to Shenana et al., (2007) and El-Alfy et al., (2018).

**Conclusion**

From such study it could be conclude that different types of wpc can be used successfully in manufacture of functional yoghurt with good physical properties that bear resemblance to that of full-fat yoghurt. Moreover; improving the sensorial qualities and physico-chemical characteristics of the produced functional yoghurt.
### Table 6. Sensory evaluation of functional yoghurt with different types of WPC when fresh and during storage periods up to 21 days at ~5°C.

<table>
<thead>
<tr>
<th>Storage periods (day’s)</th>
<th>C1</th>
<th>C2</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
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<tr>
<td><strong>Flavour (50)</strong></td>
<td></td>
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<td><strong>Appearance (10)</strong></td>
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<td><strong>Total score (100)</strong></td>
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C1: Control (full-fat standardized mixed milk cows and buffalos (1:1) ~ 3%fat).
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8