Novel probiotic camel milk yoghurt supplemented with inulin: antibacterial, antioxidant and antidiabetic effects

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Abstract

Similar to cow’s milk, camel milk also contains the essential nutrients as well as potentially therapeutic compounds with antihypertensive, and antioxidant properties. In the current study, camel milk was used for developing a new probiotic camel milk yoghurt with commercial prebiotic (inulin). The camel yoghurt samples were evaluated by monitoring the changes in some physicochemical properties (pH, total acidity and synerisis) and bacterial viability and survival of Limosilactobacillus fermentum CABA16 during storage. Besides, antibacterial, α-amylase and α-glucosidase inhibitions and antioxidant activities and the sensory evaluation of this novel product were assessed. The pH values of samples decreased whereas total acidity values increased throughout 21 days of storage and Limosilactobacillus fermentum CABA16 maintained good viability with counts higher than 10^7 CFU g^-1 at the end of storage. α-Glucosidase and α-Amylase Inhibitions Water-Soluble Extracts were higher than 35 and 55 % respectively in probiotic (PY), prebiotic (InY) and symbiotic (SY) camel yoghurts at the end of storage period. Moreover, the highest antioxidant activities of the WSEs from camel yoghurts were around 49 and 61 % by DPPH and ABTS assays respectively. The fortified probiotic camel yoghurts exhibited comparable antibacterial activities with maximum diameter of 12±0.07 mm on Staphylococcus aureus and Listeria monocytogenes strains. Lastly, the addition of probiotic and inulin significantly improved the sensory characteristics, except the colour, of camel yoghurts.

Key words: camel milk; probiotic bacteria; inulin; antibacterial; antidiabetic; antioxidant
Introduction

The concept of functional foods has led to the development of varieties of food products that not only provide basic nutrients, but can also provide benefits for human health (Gouda et al., 2021; Fazilah et al., 2018; Ozcan and Karaman, 2021).

Dairy products are known for their therapeutic characteristics, including physiological and nutritional functions. These are sources of vitamins, such as A, B, D and E, and various minerals (Mostafai et al., 2019). Yoghurt is considered one of the prevalent choices and considered as a functional fermented milk product for being a good source of essential nutrients (Mudgil et al., 2018; Tomar et al., 2021). Basically, yoghurt can be classified into two groups, standard yoghurt produced from the standard starter cultures and bio-yoghurt or probiotic yoghurt, which is supplemented with probiotic cultures (Pandey et al., 2017). Probiotic yoghurt, when consumed in adequate levels (at least 10^6 CFU mL^-1) may provide benefit effects for human health such as reduction of diarrhoea and lactose intolerance symptoms, control the serum cholesterol level due to anti-hypertensive agents (Ngongang et al., 2016) and reducing risk of colon cancers and diabetes (Barengolts et al., 2019; Rea et al., 2018).

During fermentation, the probiotics play an important role in assuring the preservation of milk by producing lactic acid and antimicrobial compounds even against viruses such as SARS-CoV-19 (Gouda et al., 2021). In this environment, most of fermented dairy products are produced from cow milk. Nevertheless, it is important to use alternative milk sources for dairy products.

Camels are found in Africa and Asia, and are kept mostly by nomads and tribes living in the arid regions. The worldwide production of camel milk is in progress which was around 2700 tons of which 1092 tons were produced in Tunisia (FAO, 2019). Nowadays, camel milk production is in progress in the regions where climatic conditions make it difficult to produce bovine milk. Camel milk has specific properties like its long shelf life without heat treatment compared to cow’s milk.

The health-beneficial importance of camel milk in the human diet has been taken into account (Kaskous, 2016; Izadi et al., 2019). In addition to its higher content of essential fatty acids, camel milk contains antimicrobial agents against gram-positive and gram-negative microorganisms thus increasing its stability over time (Kumar et al., 2015). Moreover, due to the high content of antioxidant and antimicrobial compounds, camel milk supports a vital, healthy functions of the body, such as immunoglobulin, lactoferrin, and lysozyme (Dharmisthaben et al., 2021). Also, many studies highlighted that the bioactive peptides derived from proteins such as αs1, αs2, and β-caseins play the important role in the antioxidant potential of camel milk (Izadi et al., 2019).

Several dairy products made from camel milk, including various traditionally fermented products such as koumiss, pasteurized camel milk, cheese and ice-cream have been developed and sold in many countries (Ayyash et al., 2020). Nutraceutical properties of the fermented camel milk products have been taken into consideration in some research and review articles (Ayyash et al., 2017; Ayyash et al., 2020).

Symbiotic dairy products fortified with prebiotics and probiotics continue to increase as consumers look for favourable foods that fulfil their health needs (Balthazar et al., 2019). Prebiotics belong to the category of functional food and can be defined as the non-digestible food ingredients that beneficially affect their host by selectively providing a positive influence on the probiotic bacteria growth or activity in the colon, preventing constipation, lowering blood cholesterol and improving body defences (Balthazar et al., 2017).

Inulin is a natural prebiotic ingredient which is a mixture of fructooligo- and polysaccharides and recognized functional effects in human health. This fibre also has technological characteristics without changing sensory properties and increasingly used in many food (Heydari et al., 2017). Several studies reported that inulin was able to stabilise dairy products by the formation of soluble protein-polysaccharide complexes and the increase of viscosity (Yu et al., 2021). To the best of our knowledge, no studies are carried out to investigate in-vitro the health promoting potential of camel yoghurt (antibacterial, anti-diabetic and antioxidant activities) prepared by addition of *Limosilactobacillus fermentum* probiotic strain.

Considering the health claims of the functional dairy market and the opportunity to advance the technology of dairy foods based on camel milk with functional characteristics, the current study attempted to highlight a new functional camel milk yoghurt and assessing the physicochemical characteristics, the functional properties including antibacterial, α-amylase and α-glucosidase inhibitions and antioxidant activities and the sensory evaluation of this novel product during 21 days of storage at 4 °C.

Materials and methods

Media, chemicals and probiotic growth conditions

Gelatin and date syrup were purchased from a local supermarket. MRS (de Man Rogosa and Sharpe) broth and agar (BioKar Diagnostics, France)-Vancomycin (Sigma-Aldrich, France) (20 mg L^-1) were used for the selective enumeration of *Limosilactobacillus fermentum* CABA16 under anaerobic conditions at 37 °C for 48 h (Coeuret et al., 2003). For the enumeration of *Lactobacillus delbrueckii* ssp. *bulgaricus*, MRS Agar (BioKar Diagnostics, France) was used and the plates were incubated anaerobic conditions at 37 °C for 48 h. M17 Agar (BioKar Diagnostics, France) was used for enumeration of *Streptococcus thermophilus* by incubating aerobically at 44 °C for 48 h. The probiotic cells...
of *Limosilactobacillus fermentum* CABA16 were prepared by cultivating in 100 mL MRS Broth at 37 °C for 24 h under anaerobic conditions. The culture was then centrifuged (10,000 rpm, 15 min, 4 °C) and the cells were washed twice and reconstituted in Phosphate Buffered Saline (PBS) (Sigma, France). They were then served as inoculum for production of probiotic camel yoghurts.

**Camel yoghurt processing**

Yoghurt made of camel milk was produced according to Al-Nabulsi et al. (2015) with minor modifications. Camel milk used in this study was collected from reared camels (*Camelus dromedarius*) from a local farm in the south Tunisia. Briefly, milk samples were pasteurized at 90 °C for 10 min in a water bath for whey protein denaturation followed by cooling at 43 °C. The milk samples were then inoculated with 1 % of commercial yoghurt culture (Chr. Hansen, Denmark) 10 % date syrup and 0.5 % bovine gelatin (Mudgilet al., 2018). Overall, four different formulations added with or without inulin or probiotic were prepared as follows: 1) a control sample inoculated with standard cultures (CY); 2) a yoghurt inoculated with standard cultures and probiotic culture (10⁶ CFU mL⁻¹) (PY); 3) a yoghurt inoculated with standard cultures and inulin (5 % w/v) (InY) (Heydari et al., 2017); 4) a symbiotic sample inoculated with standard cultures, probiotic and inulin (SY) followed by mixing for 1 min. Camel milk samples were fermented in an oven at 43 ± 1 °C. The fermentation was interrupted when the pH reached 4.5. Finally, the samples were packaged in propylene containers (200 mL) and stored at 4 °C for 21 days. Camel milk yoghurts were sampled at 1, 7, 14, and 21 days of storage.

**Physico-chemical analysis**

Total acidity expressed as lactic acid percentage was determined by titrating camel yoghurt samples with 0.01 NNaOH (AOAC, 2012). The pH values of yoghurt samples were detected using a Microprocessor pH-meter BT-500 (Boeco, Hamburg, Germany). Then, the syneresis of camel yoghurts were determined as recommended by (AOAC, 2012). Briefly, 10 mL of yoghurt was centrifuged (10,000 rpm, 12 min, 4 °C) and the supernatant was recovered and weighed, thereafter, syneresis was calculated as follows:

\[
\text{Syneresis} = \frac{W_1}{W_2} \times 100
\]  

Where: \( W_1 \) = Weight of supernatant and \( W_2 \) = Weight of camel yoghurt sample.

**Water-soluble extract**

For each sample, 10 g was mixed with 50 mL of 10 % Phosphate Buffer Saline. After that, the mixture was collected by centrifugation (10000 rpm, 5 min, 4 °C) and then kept at 45 °C in shaking water bath for 1 h. Also, the mixture was collected (10000 rpm, 15 min, 4 °C) then filtered using Whatman filter paper and the supernatant was stored at -20 °C (Van Ba et al., 2017). For each assay, the Water Soluble Extracts (WSEs) were vortexed for 1 min followed by centrifugation at 10000 rpm for 5 min.

**α-Amylase inhibition assay**

The α-amylase inhibition assay was carried out according to the method described by Kim et al. (2004). The inhibition percentage was determined by measuring absorbance at 540 nm and calculated as follows:

\[
\text{Inhibition} \% = \left(1 - \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}}\right) \times 100
\]  

**α-Glucosidase inhibition assay**

The α-Glucosidase inhibition assay was carried out according to the method detailed by Kim et al. (2004) with some modifications. The inhibition percentage was calculated as follows:

\[
\text{Inhibition} \% = \left(1 - \frac{\text{Abs sample} - \text{Abs blank}}{\text{Abs control}}\right) \times 100
\]  

**Antioxidant activity**

Radical Scavenging Rate by DPPH Assay. The determination of radical scavenging activity by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay was performed according to Elfahri et al. (2016). The radical-scavenging activity was expressed as follows:

\[
\text{Scavenging rate} \% = \left(1 - \frac{\text{Abs blank} - \text{Abs sample}}{\text{Abs blank}}\right) \times 100
\]  

**Antibacterial activity**

Antibacterial ability of WSEs against pathogens such as *Salmonella typhimurium* (ATCC 25922), *Staphylococcus aureus* (ATCC 25923), *Listeria monocytogenes* (ATTC 070 101 121) and *Escherichia coli* (DHS alpha, Institute Pasteur of Tunisia) was determined using agar disc diffusion method previously described by Mushqat et al. (2019). Briefly, 0.1 mL of each overnight pathogen was spotted on soft nutrient agar (Biokar Diagnostics, France) and incubated at 37 °C for 24 h.
From each WSE, 100 μL was put on sterile discs and plates were then incubated for 24 h at 37 °C. Diameter of clear zone around discs was measured in millimetres (disc diameter included) as its antibacterial activity.

Sensory evaluation

The assessments of colour and appearance, texture, flavour, aroma, taste and overall acceptability for all camel yoghurt samples were performed by a trained panel of 30 members using nine-point hedonic scale at 7 day of the storage (9 = extremely like, 8 = very much like, 7 = moderately like, 6 = slightly like, 5 = neither like nor dislike, 4 = slightly dislike, 3 = moderately dislike, 2 = very much dislike, and 1 = extremely dislike) (Mudgil et al., 2018).

Statistical analysis

Each sample was analysed in triplicate and the figures were averaged. SPSS statistics 20.0 commercial software was used to perform statistical analysis of the results (ANOVA) (Meyners and Hasted, 2021). Tukey test (p<0.05) significance level was performed to determine significant differences between the means. Data are presented as mean ± standard deviation. The experiments were carried out in triplicate.

Results and discussion

Acidification profile and bacterial growth

The viability of the lactic acid bacteria in different camel yoghurt formulations during refrigerated storage at day 1, 7, 14 and 21 is shown in Figure 1.

During the storage time, Streptococcus thermophilus counts were ranged, as an average, from 8.3±0.08 to 9.5±0.065 log CFUg⁻¹ (Figure 1A).

At the first day of cold storage, Streptococcus thermophilus counts varied from 8.3±0.08 to 8.5±0.02 log CFUg⁻¹ in CY, PY, InY and SY respectively (p<0.05). On day 14, the counts of Streptococcus thermophilus were significantly different (p<0.05). After 14 days of storage time of camel yoghurts, Streptococcus thermophilus counts decreased but it was not statistically significant (p<0.05).

Similar trends were observed during shelf-life in the counts of Lactobacillus delbrueckii ssp. bulgaricus (Figure 1B). After one week of cold storage, Lactobacillus delbrueckii ssp. bulgaricus counts varied from 8.7±0.1 to 9.1±0.14 log CFUg⁻¹. Numbers of Lactobacillus delbrueckii ssp. bulgaricus raised in the second week of storage to approximately 9.2±0.05 log CFUg⁻¹ in symbiotic camel yoghurt sample (SY) with significant difference compared to the control one (CY) (p<0.05). The addition of inulin (5%) and inoculation of probiotic bacteria did not statistically influence the Lactobacillus delbrueckii ssp. bulgaricus counts throughout the storage period in yoghurt samples, except at day 14. The bacteria counts were in agreement with observation of Alina and Kayanush (2020) and Demirci et al. (2020) who found similar trends of Streptococcus thermophilus and Lactobacillus delbrueckii ssp. bulgaricus counts in camel yoghurt and yoghurt fortified with tomato powder respectively and reported that probiotics and tomato powder did not generate any statistically considerable influence on fermentations yoghurt numbers.

Values are means±SD of n=3. CY: Control (Yoghurt) sample; PY: Yoghurt sample inoculated with added probiotic; InY: Yoghurt sample inoculated with added Inulin as prebiotic; SY: Yoghurt sample inoculated with added probiotic and Inulin as prebiotic. Lower-case letters show the differences between the samples in the same storage times and upper-case letters indicate differences between the storage times of samples (p<0.05).

Figure 1. Microbial changes of camel yoghurts inoculated with Streptococcus thermophilus (A), Lactobacillus delbrueckii ssp. bulgaricus (B), and Limosilactobacillus fermentum (C), during storage at 4 °C.
Table 1. Changes in pH, total acidity and syneresis of fermented camel yoghurts

| Storage time (days) | CY              | PY          | InY          | SY          |
|---------------------|-----------------|-------------|--------------|-------------|
| pH                  |                 |             |              |             |
| 1                   | 4.57 ± 0.14(A, A) | 4.55 ± 0.14(B, A) | 4.55 ± 0.014(B, A) | 4.53 ± 0.013(B, A) |
| 7                   | 4.5 ± 0.11(B, A) | 4.5 ± 0.19(B, A) | 4.44 ± 0.018(B, A) | 4.5 ± 0.018(B, A) |
| 14                  | 4.31 ± 0.1(B)   | 4.47 ± 0.011(B, A) | 4.45 ± 0.011(B, A) | 4.49 ± 0.11(B, A) |
| 21                  | 4.27 ± 0.022(B, A) | 4.41 ± 0.01(B, A) | 4.41 ± 0.1(B) | 4.41 ± 0.01(B, A) |
| Total acidity (%)   |                 |             |              |             |
| 1                   | 0.11 ± 0.022(A, A) | 0.10 ± 0.0022(A, A) | 0.11 ± 0.022(A, A) | 0.10 ± 0.0022(A, A) |
| 7                   | 0.15 ± 0.031(A, B) | 0.10 ± 0.031(A, A) | 0.12 ± 0.031(B, A) | 0.10 ± 0.031(A, B) |
| 14                  | 0.22 ± 0.011(B, C) | 0.18 ± 0.0011(B, C) | 0.20 ± 0.011(B, C) | 0.17 ± 0.0011(B, C) |
| 21                  | 0.35 ± 0.02(B, C) | 0.30 ± 0.002(B, C) | 0.34 ± 0.02(B, C) | 0.25 ± 0.002(B, C) |
| Syneresis (%)       |                 |             |              |             |
| 1                   | 16 ± 0.011(A, A) | 16 ± 0.001(A, A) | 16 ± 0.001(A, A) | 16 ± 0.001(A, A) |
| 7                   | 18 ± 0.0011(B, A) | 18 ± 0.0011(B, A) | 18 ± 0.0011(B, A) | 16.5 ± 0.001(A, A) |
| 14                  | 20 ± 0.011(B, C) | 18.5 ± 0.0011(B, C) | 18.5 ± 0.001(B, C) | 17 ± 0.001(A, C) |
| 21                  | 24 ± 0.022(B, D) | 21 ± 0.022(B, C) | 23 ± 0.022(B, D) | 20 ± 0.022(B, B) |

*Values are mean ± SD (n = 3). Legend: CY: Control (Yoghurt) sample; PY: Yoghurt sample inoculated with added probiotic; InY: Yoghurt sample inoculated with added prebiotic and probiotic; Lower-case letters show the differences between the samples in the same storage time and upper-case letters indicate differences between the storage times of samples (p<0.05).
Antibacterial and antioxidant activities

The inhibition ability of the WSEs against pathogens is summarized in Table 2.

The results show that the WSEs inhibited the growth of Listeria monocytogenes, Staphylococcus aureus, Salmonella Typhimurium and Escherichia coli. Antibacterial activities of WSEs were significantly higher than control (p<0.05) with a maximum inhibition diameter of 12±0.07 mm exhibited by symbiotic camel yoghurt against Staphylococcus aureus. The antibacterial activities of PY, InY and SY against all tested pathogens were not significant (p>0.05).

The antibacterial behaviour of fermented camel milk samples may be attributed to several factors such as the biofunctional properties of camel milk proteins including lysozyme, lactoferrin, and immunoglobulin, reduction of camel yoghurt pH, influence of storage temperature, and the production of antimicrobial compounds such as bacteriocins and peroxide hydrogen besides lactic acid by LAB including probiotics (Izadi et al., 2019).

Conesa et al. (2008) evaluated the antimicrobial potential of lactoferrine isolated from goat, sheep, camel, and human milks against Salmonella typhimurium and Escherichia coli 0157:H7. Results showed that lactoferrin from camel milk had the maximum antibacterial activity against Escherichia coli 0157:H7. Also, Benkerroum et al. (2004) reported that Listeria monocytogenes and E. coli could be inhibited in camel milk at 4 °C by Lactoperoxidase system and lysozyme. Al-Nabulsi et al. (2015) showed that Listeria monocytogenes number was reduced in camel yoghurt during refrigerated storage. Jrad et al. (2015) also explored that the growth number of Escherichia coli could be reduced by 19.3 % in the presence of 20 g/L camel milk casein. Recently, Algboony and Muhialdin (2021) have realized that the fermented camel milk with Lactobacillus plantarum could significantly inhibit the cell growth of Escherichia coli 0157:H7 and Staphylococcus aureus. Camel milk contains significantly higher concentrations of whey proteins, including lysozyme, lactoferrine and IgG which are markedly more heat resistant than their in cow milk, although the heat treatment may destroy part of the whey proteins in camel milk. Finally, the inoculated probiotic bacteria enhanced the antimicrobial activity of camel milk.

Furthermore, symbiotics have great antimicrobial properties (Cadieux et al., 2008). The combination of probiotic bacteria with prebiotic compound can cause the release of antibacterial substances such as bacteriocins, which can reduce the growth of pathogens (Faziliah et al., 2018).

Figure 2. α-glucosidase (A) and α-Amylase (B) inhibition of camel yoghurts

The inhibitory effects of camel yoghurt formulations on α-glucosidase and α-amylase activities as indicators for antidiabetic activities through 21 days of storage at 4 °C are presented in Figure 2A and B, respectively.

Noteworthy, the inhibition of α-glucosidase and α-amylase increased (p<0.05) with storage period. The percentages of α-glucosidase inhibition increased during storage and ranged from 27±0.08 to 44±0.08 % (Figure 2A). The probiotic, prebiotic and symbiotic WSEs exhibited significant α-glucosidase inhibition compared to the control during two weeks of storage (p<0.05). Similarly, α-amylase inhibitions were significantly greater (p<0.05) in PY, InY and SY samples than control during storage (Figure 2B). The PY and SY camel yoghurts showed higher α-amylase inhibition activity with final percentages of 58±2.5 and 57±0.5 % respectively.

Table 2. Antibacterial activity expressed as inhibition zone diameter (mm) of camel yoghurts

| Pathogenic bacteria          | Camel yoghurt samples | CY       | PY       | InY       | SY       |
|------------------------------|-----------------------|----------|----------|-----------|----------|
| Listeria monocytogenes      | 4±0.01<sup>±</sup>    | 11.8±0.05<sup>a</sup> | 11.2±0.007<sup>b</sup> | 12±0.007<sup>b</sup> |
| Staphylococcus aureus       | 4.9±0.015<sup>a</sup> | 12±0.011<sup>b</sup> | 11±0.07<sup>b</sup>  | 12±0.07<sup>b</sup>  |
| Salmonella Typhimurium      | 3.5±0.001<sup>a</sup> | 8.8±0.002<sup>b</sup> | 7.6±0.01<sup>b</sup> | 8±0.01<sup>b</sup>   |
| Escherichia coli            | 3.1±0.01<sup>a</sup>  | 8±0.007<sup>b</sup>  | 7.5±0.019<sup>b</sup>| 8.5±0.019<sup>b</sup>|

*Values are mean ± SD (n=3); Legend: CY: Control (Yoghurt) sample; PY: Yoghurt sample inoculated with added probiotic; InY: Yoghurt sample inoculated with added prebiotic; SY: Yoghurt sample inoculated with added probiotic and prebiotic; Different letters (a-c) present the differences between the samples in the same storage time (p<0.05)
The antidiabetic activities of different treatments were similar to those of Ayyash et al. (2017) who reported that changes in α-amylase and α-glucosidase inhibitions were important in fermented probiotic fermented camel milks. Similarly, Balthazar et al. (2019) showed that the probiotic sheep milk juice containing both fibres inulin or potato starch showed higher inhibition of digestive enzymes. The noticeable α-glucosidase inhibitions may be attributed to the bioactive peptides released by the proteolytic action of probiotics in camel yoghurts (Ayyash et al., 2018). Similarly, prebiotic addition (inulin) enhanced the proteolytic activity of this bacteria, providing more bioactive peptides in the functional camel yoghurts. Moreover, camel milk directly affects the insulin receptor function and glucose transport in the body’s insulin-sensitive tissues to regulate glucose homeostasis (Izadi et al., 2019). Overall, the high enzyme inhibitory potential of camel milk may be owing to the release of small bioactive peptides as a result of the secretion of proteolytic enzymes by probiotic strains (Kumar et al., 2016).

The DPPH and ABTS radical scavenging abilities of different camel yoghurt formulations were shown in Figure 3. DPPH radical scavenging ability increased during storage period (p<0.05) in all samples. DPPH inhibition values of probiotic (PY) and inulin (InY) enriched yoghurts and their combination (SY) were 48±2.4, 44±2.5 and 49±1.5 % respectively when this value was 39±1.7 % for control yoghurt (CY) after 21 days of refrigerated storage (p<0.05). In addition, ABTS radical scavenging ability of camel yoghurt samples also increased to reach a maximum of 61±1.3 % in symbiotic yoghurt. It can be seen from results that PY, InY and SY are more effective than CY for both DPPH and ABTS radical scavenging.

Antioxidant activity inhibits the oxidation of molecules caused by free radicals and is important for the shelf life of dairy foods and to protect the human body against oxidative damage upon consumption (Izadi et al., 2019). Our results are in same line as previous study demonstrated that inulin enhanced the proteolytic activity of Lp B2, producing bioactive peptides (Balthazar et al., 2019). This could be explained by crucial role of bioactive compounds in foods, in elevating the effect of reactive oxygen species such as superoxide, hydroxyl, and peroxyl radicals formed by cells under oxidative stress (Dharmisthaben et al., 2021).

These bioactive compounds, especially protein derived peptides, can donate electrons to neutralize free radicals. Moreover, the presence of several amino acid residues in the peptide chains can enhance antioxidant properties. Moreover, the presence of high amounts of antioxidant enzymes and non-enzymatic antioxidants (e.g., glutathione, and vitamins E and C) in camel milk can notably improve its antioxidant and antiradical activities, contributing to the control of tissue damage (Shori, 2015). Nonetheless, bioactive peptides derived from camel milk proteins (mainly α1-, α2-, and B-caseins) play the highest role in the antioxidant potential (Izadi et al., 2019; Oussaief et al., 2021). In consistence to our obtained results, many studies have recently interested on the fermentation role by some probiotic bacteria (include Lactobacillus rhamnosus, Lactobacillus plantarum, Lactobacillus kefiri, Leuconostoc and Streptococcus thermophilus, Lactobacillus acidophilus or Lactobacillus helveticus) in camel milk digestion to increase the antioxidant activity (Alhaj et al., 2017).

Inulin addition to camel yoghurts (InY and SY) significantly increased the antioxidant activity during storage compared with control (CY), showing a possible synergism between probiotic strain Limosilactobacillus fermentum CABA16 and inulin.

**Sensory characteristics of camel milk yoghurt**

Sensory evaluation of camel yoghurts was carried out at day 7 on a 9-point hedonic scale for colour and appearance, texture, flavour, aroma, taste and overall acceptability. Treatments have significant effects on the overall sensory parameters of camel yoghurt samples (p<0.05). Inoculation of probiotic bacteria (Limosilactobacillus fermentum CABA16) and the addition of prebiotic (inulin) had a great significance on the texture, aroma, taste, flavour, appearance and overall acceptability of camel yoghurts (p<0.05) (Figure 4). Whereas, colour was not affected by the addition of both probiotic and prebiotic. Maximum score of texture was found in the SY sample (6±0.24). Therefore, minimum scores were obtained by control sample (CY).
The results are in same line as per previous findings of (Mohsin et al., 2019) who showed that the sensory characteristics of camel yoghurt was enhanced by biosynthesized xanthan. Moreover, Yu et al. (2021) reported that yoghurt sample fortified with inulin showed highest acceptance, flavour and texture scores compared with the control yoghurt.

Conclusions

In the present study a new functional fermented camel milk fortified with Limosilactobacillus fermentum CABA16 and inulin as prebiotic has been developed. Limosilactobacillus fermentum CABA16 number remained up to 8 log_{10} CFUg^{-1} during 21 days of refrigerated storage. In addition, the probiotic culture improved the physico-chemical and sensory characteristics, the antibacterial, antioxidant and antidiabetic activities of the camel yoghurt, especially when combined with inulin. Finally, the obtained results contribute to the scientific support to the process camel milk into dairy products. The presented results also encourage further investigations into the assessment of other functional properties and characterisation of the bioactive peptides derived from probiotic camel yoghurt.

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Probiotički jogurt od devinog mlijeka s dodatkom inulina: antibakterijsko, antioksidacijsko i antidiabetičko djelovanje

Sažetak

Devno mlijeko poput kravljeg sadrži esencijalne hranjive tvari kao i potencijalno korisne spojeve s antihipertenzivnim i antioksidativnim svojstvima. U ovom je istraživanju korišteno devino mlijeko kao sirovina za razvoj novog probiotičkog jogurta s komercijalnim prebiotikom (inulin). U uzorcima devnog jogurta tijekom skladištenja prateone su promjene nekih fizikalno-kemijskih svojstava (pH, ukupna kiselost i sinereza) te preživljavanje bakterija uključujući i soj Limosilactobacillus fermentum CABA16. Osim toga, proizvedeni jogurti su senzorski ocijenjeni te im je određena antibakterijska i antioksidacijska aktivnost, kao i sposobnost inhibicije α-amilaze i β-glukozidaze. Tijekom 21 dana skladištenja, pH vrijednosti uzoraka su padale, dok su vrijednosti ukupne kiselosti porasle, a soj Limosilactobacillus fermentum CABA16 pokazao je dobru sposobnost preživljavanja budući je na kraju skladištenja broj poraslih kolonija iznosio preko 10^7 CFU g^{-1}. Inhibicije α-glukozidaze i α-amilaze u ekstraktima topljivim u vodi bile su veće od 35 odnosno 55 % u probiotičkom (PY), prebiotičkom (InY) i simbiotičkom (SY) devnom jogurtu na kraju razdoblja skladištenja. Štoviše, najveće antioksidacijske aktivnosti ekstrakta topljivog u vodi (WSE) dobivenog obradom uzoraka devnih jogurta bile su oko 49 odnosno 61 % prema DPPH i ABTS testovima. Obogaćeni probiotički devni jogurti pokazali su slično antibakterijsko djelovanje na sojeve Staphylococcus aureus i Listeria monocytogenes, s maksimalnim promjerom zona inhibicije od 12±0,07 mm. U konačnici, dodatak probiotika i inulina značajno je poboljšao sva senzorska svojstva devnih jogurta osim boje.

Ključne riječi: devio mlijeko; probiotičke bakterije; inulin; antibakterijski; antijabestik; antioksidans
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