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Microbiological, Biochemical, and Organoleptic Properties of Probiotic Dairy Desserts Made from Camel Milk and *Echinophora platyloba* Extract Nanoemulsion

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Abstract

This study aimed to produce *Lactobacillus Plantarum* bacteria and the *Echinophora platyloba* extract nanoemulsions by sodium alginate to enrich dairy dessert based on camel milk. For this, the amount of 0.6, 1.25, and 2.5% *Echinophora platyloba* extract was added to dessert. Extract nanoemulsion, dairy desserts physicochemical, microbial, and overall acceptance was tested. The viability of probiotic bacteria was performed on 1, 11, and 21 days in three replications. The results showed the formation of fine and uniform particles. The nanoemulsion of the extract caused more preservation of phenolic compounds and thus increased their antioxidants. The diffusion result shows that *Escherichia coli* and *Candida albicans* were the most resistant and sensitive microorganisms, respectively. The nanoemulsion of the extract showed the MIC and MBC on *Escherichia coli*, *Candida albicans*, and *Aspergillus niger*. However, no statistically significant difference was observed between the MIC of the free and nanoemulsion extract on *Staphylococcus aureus*. Dairy desserts test showed an increase in acidity, ash and overall acceptance, a decrease in pH and moisture with an increase in the concentration of the fine-grained extract, and the storage time. The used extract had no significant effect on the amount of fat and protein in the treatments. The microbial test showed that bacteria did not grow against the microencapsulated extract. An increase in the viability of probiotic bacteria in dairy desserts was also reported. Therefore, dessert milk based on camel milk can be introduced as a beneficial probiotic product.

Keywords: Camel milk, Dairy dessert, *Echinophora platyloba* extract, *Lactobacillus Plantarum*, Microencapsulation

Introduction

Recently, dairy desserts represent a great potential to produce probiotic cultures. Probiotics are represented by the International Scientific Association for Probiotics and Prebiotics as live microorganisms that deliberate a health benefit when administered in suitable amounts (Guimarães *et al.*, 2020). One of the probiotics is *Lactobacillus Plantarum* (Hanafi *et al.*, 2022). Even though the probiotic microorganisms conduct to remain after processing and during storage, some of them are killed when exposed to the difficult condition of the upper digestive tract. Hence, the preservation of living probiotic microorganisms became a severe interest. Probiotics are preserved, in an encapsulation system to prevent the microorganisms from damage, increase shelf storage life, and provide adequate protection from the acidic

environment (Pimentel *et al.*, 2021). Microencapsulation of microorganisms and biologically active substances in various foods produce considerable possibilities to obtain products with increased biological value and potential health effects. Encapsulation of *Echinophora platyloba*, rich in polyphenolic compounds was used to obtain a new fermented dairy dessert (Nasiri *et al.*, 2021).

Echinophora platyloba is a perennial herbaceous plant of the genus *Echinophora* (30-100 cm height) covering the short, curved trichome and a hard, thick stem. Previous studies have shown that this plant contains saponins, flavonoids, and alkaloids, and one of the most critical terpene compounds identified in it includes trans-beta- ocimene with a concentration of 67.9% (Mahboubi, 2009). The antibacterial effects associated with its extract on *Staphylococcus aureus* and *Streptococcus faecalis* have also been proven (Avijgan *et al.*, 2006). Until now no available information about the encapsulation of *Echinophora platyloba* extract has been reported to be applied in dairy desserts from camel milk. The aim of the current study is to investigate the Microbiological, biochemical, and organoleptic properties of probiotic dairy desserts made from camel milk and enriched with *Echinophora platyloba* extract nanoemulsion to improve their health and functional characteristics.

Materials and methods

To prepare EPE, the dried herb of *Echinophora platyloba* was purchased from a reputable grocery in Khorasan (Iran). The extraction was then performed by soaking and concentrating the extract with a rotary apparatus connected to a vacuum pump and finally nanoemulsion of extract was prepared by sodium alginate (Marsup *et al.*, 2020). In this study, the amount of 0.6, 1.25, and 2.5% *Echinophora platyloba* extract nanoemulsions were added to the dessert.

The probiotic culture *Lactobacillus planturum* (Lp), in freeze-dried form was provided from Chr. Hansen company (Denmark). The nanoemulsion technique was used for the preparation of *Lactobacillus Plantarum* bacteria according to the method described by (Marsup *et al.*, 2020). For dairy desert preparation, the following materials were used according to the Seuvre *et al.* (2008) method:

- Camel milk was purchased from a reputable grocery in Gorgan (Iran), with 4.91% fat, 2.9% protein, 4.52% lactose, 0.77% ash and 0.21% acidity.
- The amount of 0.6, 1.25, and 2.5% *Echinophora platyloba* extract nanoemulsions.
- 1 g probiotic culture *Lactobacillus planturum* nanoemulsions.
- Sugar
- Gelatin
- Malt

Minimum inhibitory concentration (MIC) and Minimum bactericidal concentration (MBC) is done by microdilution method according to (El Hamdaoui *et al.*, 2018). MIC and MBC was determined for free and nanoemulsion extract on *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, and *Aspergillus niger*.

Chemical analysis such as: Acidity and pH contents of individual dessert samples were measured according to Seuvre *et al.*, 2008 method. Fat, protein, moisture and ash of probiotic samples was determined according to Iranian National Standardization Organization [ISIRI] (2012). Microbial test which was performed according to Iranian National Standardization Organization [ISIRI], (2014). For survival of *L. Plantarum*, this probiotic bacteria was grown in the MRS Bile Agar and incubated at 37 °C for 72 h. Survival of the probiotic was determined following 1, 11, and 21 days of storage at 4 °C.

For measuring the total polyphenol content and antioxidant properties: Rezaei Savadkouhi *et al.* (2020) method was employed to estimate the total phenolic content of dessert samples. Percentage of DPPH free radical scavenging activity (2,2-diphenyl-1-picrylhydrazyl) was performed using Rezaei Savadkouhi *et al.* (2020) method.

For sensory analysis, ten skilled panellists were selected to complete the sensory analysis. The scores were reported based on the Hedonic 5-point numbers. For statistical analysis, all results presented in this study are the means of three independent experiments with three replicates each. Data were analysed using one-way analysis of variance (ANOVA) with SPSS software.

Results and discussion

The results of total phenol content and antioxidant properties revealed that *Echinophora platyloba* extract nanoemulsion had higher total phenol content than *Echinophora platyloba* extract samples. Also, the antioxidant properties of nanoemulsion samples were higher than in samples without extract nanoemulsion. In the current study, our findings demonstrated that nanoemulsions by sodium alginate protect the extract and lead to an increase in the total phenol content and antioxidant properties of samples.

The diffusion result shows that *Escherichia coli* and *Candida albicans* were the most resistant and sensitive microorganisms, respectively. The nanoemulsion of the extract showed a significant difference for MIC and MBC on *Escherichia coli*, *Candida albicans*, and *Aspergillus niger*. However, no statistically significant difference was observed between the MIC of the free and nanoemulsion extract on *Staphylococcus aureus*.

The results of the chemical analysis evaluated that the changes in titratable acidity of the samples during the 21 days of storage period indicated that the samples containing EPE had a significant difference from the control sample ($P \leq 0.05$). The highest increase in average acidity was observed at the end of the storage period. The sample containing 2.5% EPE consistently had the highest acidity during the storage period, with an average of 0.35 ± 0.001 acid lactic. The results revealed that increasing EPE concentration had decreased pH. The dessert sample containing 2.5% EPE consistently had the lowest pH during the 21 storage period, with an average of 5.52 ± 0.001 . The results revealed that the used extract had no significant effect on the amount of fat and protein in the treatments. The results of evaluating the changes in moisture of the samples during the 21 days of storage period indicated that the samples containing EPE had a significant difference from the control sample ($P \leq 0.05$). The moisture of the sample decreased during the 21 days of storage period and The *Echinophora platyloba* extract had a significant effect on the amount of moisture in the treatments.

The results of the microbial test showed that the samples containing EPE and the control sample had a significant difference from each other in each of the evaluation points ($P \leq 0.05$). Based on these results, the bacteria did not grow against the microencapsulated extract. Moreover, a significant increase in the viability of probiotic bacteria in dairy desserts with microencapsulated extract was also reported. The results of the organoleptic evaluation showed that EP extract had a significant increase in the level of overall acceptance in the treatments. During the 21 days of storage in probiotic dairy desserts, the overall acceptance level reduced significantly ($P < 0.01$).

Conclusions

In conclusion, the results of this study indicated that nanoemulsion of probiotic *L. plantarum* increased bacterial survival During the 21 days of days of storage. The nanoemulsion of the *Echinophora platyloba* extract caused more preservation of phenolic compounds and thus increased their antioxidants and increased acidity, ash, and overall acceptance of dairy dessert. Therefore, microencapsulation technique was useful to develop probiotic dairy

desserts from camel milk enriched with *Echinophora platyloba* extract for improving their health and functional characteristics.

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