





DEVELOPMENT AND EVALUATION OF FUNCTIONAL DAIRY FOODS AND BEVERAGES

$\mathbf{B}\mathbf{y}$

Walaa Mohamed Saad Bahnas

B.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2012)

M.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2020)

Thesis

Submitted in Partial Fulfillment of the Requirements for the Degree of

Doctor of Philosophy

In

Agricultural Sciences (Dairy Science)

Dairy Department, Faculty of Agriculture Fayoum University, Egypt

2024

Development and evaluation of functional dairy foods and beverages

By

Walaa Mohamed Saad Bahnas

B.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2012) M.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2020)

Thesis

Submitted in Partial Fulfillment of the Requirements for the Degree of

Doctor of Philosophy

In

Agricultural Sciences (Dairy Science)

Supervision Committee:

| 1. Prof. Dr. Khaled A. Abdel-Sala | ım Abbas (Main supervisor) |
|------------------------------------------|------------------------------|
| Professor of Dairy Sci., Fac. of Agric., | Fayoum University |
| Signature | |
| 2. Dr. Nesreen Mohamed Nasr | (Supervisor) |
| Assistant Professor of Dairy Sci., Fac. | of Agric., Fayoum University |

Signature

APPROVAL SHEET

Development and evaluation of functional dairy foods and beverages

By

Walaa Mohamed Saad Bahnas

B.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2012)M.Sc. Agric. Sci. (Dairy Sci.), Fac. of Agric., Fayoum Univ. (2020)

Thesis

Submitted in Partial Fulfillment of the Requirements for the Degree of

Doctor of Philosophy

In

Agricultural Sciences (Dairy Science)

This thesis for Ph.D (Dairy Science) has been approved by:

| 1. | Prof. Dr. Ali Ibrahim Ali Mansour |
|----|----------------------------------------------------------------------------------|
| | Prof. Emeritus of Dairy Sci., Fac. of Agric., Al-Azhar University, Assiut Branch |
| | Signature |
| 2. | Dr. Ratiba Bayoumi Ahmed |
| | Assistant Professor emeritus of Dairy Sci., Fac. of Agric., Fayoum Univ. |
| | Signature |
| 3. | Prof. Dr. Khaled A. Abdel-Salam Abbas (Main supervisor) |
| | Prof. of Dairy Sci., Dairy Dept., Fac. of Agric., Fayoum Univ. |
| | Signature |
| | |

Date of Examination: 4/9/2024

ABSTRACT

The main aim of this thesis was to develop and evaluate the quality of a new probiotic fermented milk beverage enriched with perilla or psyllium seeds using a new probiotic strain isolated from traditionally fermented milk products. This thesis is conducted in three main parts as follows:

In the first part: The aim was to isolate, identify and characterize new probiotic lactic acid bacteria (LAB) isolated from 15 samples of traditional fermented dairy products (Laban Rayeb, Laban Khad, and Kariesh cheese) collected from Fayoum, Egypt. 25 LAB isolates were isolated, and identified morphologically, biochemically into three groups: the genus Enterococcus/lactococcus (13 isolates), Lactobacillus (6 isolates), and Leuconostoc (6 isolates). Of the 25 isolates, 4 LAB isolates (LAB-3, LAB-4, LAB-13, and LAB-14) showed promising characteristics as probiotics by expressing acid, bile salt tolerance, antibiotic susceptibility and antimicrobial activity against the tested pathogenic bacteria. The 4 LAB isolates were further identified by 16S rDNA gene sequencing. The phylogenetic tree based on 16S rDNA indicated that isolate LAB-3 showed a 100% match with Enterococcus durans MT545074.1, LAB-4 showed 99.93% similarity with Enterococcus lactis MH348130.1, LAB-13 showed 100% homology with *Lactobacillus acidophilus* MT604714.1, and LAB-14 showed 99.70% homology with Leuconostoc mesenteroides MT538672.1. In conclusion, our isolates of Enterococcus durans isolated from traditional Egyptian dairy products showed promising characteristics as probiotics and could be effectively utilized in the development of fermented milk beverages.

In the second part, the effect of different levels of psyllium and perilla seeds (0.0, 1.0, 1.5, and 2.0%) on the viability of *Enterococcus durans* and *Lactobacillus acidophilus* in MRS broth, skim milk and gastrointestinal tract (in vitro) were investigated. The results showed that the viability of *Enterococcus duras* and *Lactobacillus acidophilus* were significantly higher when 1.5% perilla seeds were added to MRS and skim milk. Trials that achieved the highest viability will be exposed to conditions that simulate the gastrointestinal tract in vitro. The gastrointestinal simulation showed that the viability of *Enterococcus durans* and *Lactobacillus acidophilus* decreased in all samples during the gastrointestinal tract stages, but perilla or psyllium seeds provided suitable viability for probiotic bacteria compared to the control. Overall, all trials showed that the viability remained above 10⁶ cfu/g, and *Enterococcus durans* strain tested was the most promising strain and could potentially be used in fermented milk development.

In the third part, the development and quality evaluation of a new probiotic fermented milk beverage using *Ent. durans* and enriched with 1.0% psyllium, 1.0% perilla seeds, and a

mixture of 0.5% psyllium and 0.5% perilla seeds during cold storage for 14 days was evaluated. The addition of perilla or psyllium seeds to fermented milk beverage significantly increased titratable acidity, total solids, protein, fat, ash, water holding capacity, total phenolic content, antioxidant activity and viability of *Ent. durans*. Psyllium seed significantly reduced the synersis of the product. Higher antioxidant activity was observed in beverages enriched with seeds. The highest viability of *Ent. durans* was found in beverages containing 1.0% perilla or psyllium seeds. Fermented milk beverages enriched with 1.0% psyllium seeds exhibited higher acceptance from panelists during sensory evaluation.

Overall, this study highlights the potential for the development of milk beverage fermented with *Ent. durans* as a new probiotic strain enriched with 1.0% psyllium seeds to improve the functional and physiochemical properties of the product. The final product retained probiotic survival above 10⁷ cfu/g even after storage at 5°C for 14 days.

Keywords: LAB Isolation, probiotic beverages, plant ingredients, perilla seeds, psyllium seeds, *Enterococcus durans*, *Lactobacillus acidophilus*