Isolation and identification of *Lactobacillus* spp. and *Streptococcus thermophilus* towards developing probiotic Dahi

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Session: 2018-2019

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Dairy Science

Department of Dairy and Poultry Science

Faculty of Veterinary Medicine

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Chattogram-4225, Bangladesh

DECEMBER 2019

Dedicated

To My

Beloved Family

Authorization

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This is to certify that we have examined the above Master's thesis and have found that the thesis is complete and satisfactory in all respects, and that all revisions required by the thesis examination committee have been made

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Acknowledgements

I am indebted to Almighty Allah who enabled me to complete the research work and write up the dissertation successfully for the degree of Master of Science (MS) in Dairy Science under the Department of Dairy and Poultry Science (DDPS), Chattogram Veterinary and Animal Sciences University (CVASU).

I am grateful to my supervisor ***Dr. A K M Humayun Kober****, Professor and Head, DDPS, CVASU, for his valuable supervision and guidance. It was really a great pleasure and amazing experience for me to work under his supervision. I really deemed it and I realized it was a rare opportunity for me to work under his creative guidance. I understand it was impossible to complete the dissertation without his constructive supervision.*

It's my great pleasure to convey my profound gratitude to my co-supervisor ***Dr. M. A. Hossain (Rony),*** *Professor, DDPS, CVASU, for his valuable advice, scholastic guidance, suggestions and inspiration and especially for his patience and guidance during the writing process.*

It is my privilege to acknowledge ***Professor Goutam Kumar Debnath,*** *DDPS for his support, valuable advice and encouragement for the research work. Besides, I could not but thanks to all other remaining teachers (especially* ***DR. Nasima Akter****) and staff of DDPS, CVASU, for the insightful discussion, offering valuable advice, along with their valuable support, kind cooperation and help during the whole period of my study in this department.*

My special thanks go to ***DR. Eaftekhar Ahmed Rana****, Lecturer, Department of Microbiology and Veterinary Public Health, CVASU for his assistance and help during microbiological assays in microbiology laboratory.*

I would like to express my deep sense of gratitude and thanks to Vice Chancellor ***Professor Dr. Goutam Buddha Das****, Chattogram Veterinary and Animal Sciences University. My cordial thank goes to the Coordinator of Advance Studies and Research and Committee of Advance Studies and Research for providing research fund to complete my research work. I would like to acknowledge the support and co-operation received from technical and non-technical staffs of Poultry Research and Training Centre (PRTC), CVASU.*

I thank my fellow ***lab mates*** *and* ***my friend****, who have, in their own ways, kept me going on my path to success, assisting me as per their abilities, in whatever manner possible, encouraging me and for ensuring that good times keep flowing.*

Last but not the least, I express my deepest sense of gratitude to my beloved family members for their sacrifice, blessings and encouragement. I also place on record, my sense of gratitude to one and all, who directly or indirectly, have stretched out their hand in this noble work.

The Author

December 2019

Table of Contents

List of Tables

List of Figures

List of Abbreviations and symbols

Abbreviations	Elaborations
<	Less than
>	Greater than
%	Percentage
µg	Microgram
µl	Microliter
µmol	Micromole
A.	*Akkermansia*
ACE	Angiotensin converting enzyme
Amp	Ampere
ANOVA	Analysis of variance
B.	*Bifidobacterium*
BHI	Brain heart infusion
bp	Base pair
℃	Degree centigrade
Ca	Calcium
cDNA	Complementary DNA
cfu	Colony forming unit
CVASU	Chattogram Veterinary and Animal Sciences University
DDPS	Department of Dairy and Poultry Science
DM	Dry matter
DNA	Deoxyribonucleic acid
dNTP	Deoxyribonucleotide triphosphate
e.g	Example
EPS	Exopolysaccharides
et al.	And his associates
etc.	Et cetera
GDP	Gross domestic product
GIT	Gastrointestinal tract
gm	Gram

Abbreviations	Elaborations
h	Hour
HDL	High-density lipoproteins
i.e.	That is
IgE	Immunoglobin E
IU	International unit
Kb	Kilobase pair
Kg	Kilogram
L	Litre
L.	*Lactococcus*
LAB	Lactic acid bacteria
Lb.	*Lactobacillus*
LDL	Low-density lipoproteins
Ln.	*Leuconostoc*
mg	Milligram
Mg	Magnesium
min	Minute
ml	Milliliter
MRS	De Mann Rogosa Sharpe
MS	Master of Science
P.	*Propionibacterium*
PBS	Phosphate buffer saline
PCR	Polymerase chain reaction
PRTC	Poultry Research and Training Centre
rpm	Rotation per minute
S.	*Streptococcus*
SEM	Standard errors of mean
Spp.	Species
Subsp.	Subspecies
TG	Triglyceride
UV	Ultraviolate
VRB	Violet Red Bile

Abstract

Dahi claims as popular fermented dairy food product available in the consumer world. An attempt was made herein this study to develop Dahi with the specific lactic acid bacteria (LAB) isolated from locally available Dahi. For this view, local Dahi samples were collected and specific bacteria (*Lactobacillus* spp. and *Streptococcus thermophilus*) were isolated and identified by the lab analytical process. The analyses included morphological characteristics, microscopic observation and catalase reaction etc., to select the isolated bacteria, which were confirmed by Polymerase Chain Reaction-Gel Electrophoresis method. Five Dahi samples (T_1, T_2, T_3, T_4 and T_5) were prepared using the PCR identified culture. Dahi samples (T_1, T_2 and T_3) were prepared using *Lactobacillus* spp., whereas T_4 sample from *Streptococcus thermophilus* and T_5 sample was prepared by a combination of *Lactobacillus* spp. (T_3) and *Streptococcus thermophilus* (T_4) bacteria together. Physicochemical, microbiological and sensory properties were measured out to assess the quality of the developed Dahi. The shelf life of the product was assessed by determining pH and titrable acidity of the samples measured on day 1 and 7, respectively. Numerically variable pH and acidity were observed in the different samples of Dahi. There was an increase in acidity with the progress of the storage time, but the values were in acceptable range on day 7. Data on proximate analysis revealed that the ash and protein percentage of T_3 sample were significantly ($P<0.05$) better than those of other samples. The viable count of Dahi cultures was well above the minimum recommended amount (10^6 CFU/ml) in all the Dahi samples. The sensory properties namely taste, aroma and overall acceptability of Dahi prepared from *Streptococcus thermophilus* (T_4) and *Lactobacillus* spp. (T_3) were significantly ($P<0.05$) improved compared to other samples. The results suggest that the starter culture identified and utilized in this study to develop Dahi have a potential to contribute to Dahi production technology. It can be concluded that the developed Dahi in this study might be a potential dairy food item to draw the attention of the consumer demand, acceptability and consumer world for introducing a novel dairy market outlet across the globe.

Key words: Lactic acid bacteria, PCR, Probiotic, shelf life, acidity, pH, sensory characters, Dahi.