P-Edu International Journal of Multidisciplinary Studies
Volume 2, Issue 1 Jan-June (2026)
DOI: 10.70199/P-EIJMS.2.1.37-43
ORCiD: https://orcid.org/0009-0009-1328-7913

RESEARCH ARTICLE

ISSN: 3139-2830 (Online)

COMPARATIVE EVALUATION AND SELECTION OF NCDC CULTURES FOR FERMENTED SOY YOGHURT MANUFACTURE BASED ON TECHNOLOGICAL AND PROBIOTIC CHARACTERISTICS

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Article History
Received: 26 April 2026
Revised: 19 May 2026
Accepted: 04 June 2026
Published: 25 June 2026

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ABSTRACT: The selection of suitable starter and probiotic cultures is a critical factor influencing the quality, functionality and consumer acceptability of fermented soy products. The present investigation was undertaken to evaluate the technological and probiotic characteristics of six selected cultures, namely Lactobacillus acidophilus NCDC15, Lacticaseibacillus casei NCDC17, Lacticaseibacillus rhamnosus NCDC24, Lactiplantibacillus plantarum NCDC25, Bifidobacterium bifidum NCDC232 and Streptococcus thermophilus NCDC293, with the objective of identifying suitable cultures for fermented soy yoghurt manufacture. The cultures were evaluated for growth characteristics, acidification behaviour, acetaldehyde production, exopolysaccharide (EPS) synthesis, bile tolerance and cholesterol assimilation. Significant differences ($P \leq 0.05$) were observed among cultures for all evaluated parameters. NCDC293 exhibited the highest technological performance with superior growth rate (0.278 h^{-1}), shortest generation time (2.49 h), highest titratable acidity (1.12%), acetaldehyde production (30.4 ppm) and EPS production (198.2 mg/L). NCDC25 demonstrated the highest probiotic potential, recording maximum bile tolerance (94.8%) and cholesterol assimilation (61.2%). NCDC24 exhibited balanced technological and probiotic characteristics and ranked third overall according to CTPI analysis. Comparative ranking based on the Composite Technological–Probiotic Index (CTPI) identified NCDC25, NCDC293 and NCDC24 as the three most promising cultures for fermented soy yoghurt manufacture. NCDC293 exhibited superior technological performance, whereas NCDC25 demonstrated the highest probiotic potential and the highest overall CTPI score. The results suggest that the combination of Streptococcus thermophilus NCDC293 and Lactiplantibacillus plantarum NCDC25 possesses considerable potential for the development of fermented soy yoghurt with improved technological and functional properties.

Keywords: Fermented soy yoghurt, Probiotic culture, NCDC cultures, Lactic acid bacteria, Fermented soymilk, Lactiplantibacillus plantarum, Streptococcus thermophilus, Cholesterol assimilation, Bile tolerance, Exopolysaccharides (EPS), Technological-probiotic index

INTRODUCTION

As people are increasingly concerned about sustainable, healthy and eco-friendly food, the production of plant-based dairy options has surged forward. Of these, fermented plant-based foods have garnered a lot of interest as they offer the nutritional benefits of plant-based foods along with the benefits that come from microbial fermentation. The advantage of fermentation is to improve the safety of the product, digestibility, sensorial quality and shelf life, while increasing the bioavailability of nutrients and bioactive compounds [1,2]. Therefore, plant-based food products that are fermented are gaining popularity as functional foods that can provide consumers with both nutritional and physiological benefits.

Soybean (*Glycine max* L.) is one of the most valuable plant protein sources and is a main ingredient in the production of various plant-based dairy products. Soymilk is a source of high quality proteins, essential amino acids, polyunsaturated fatty acids, dietary fibre, minerals and biologically active isoflavones which have been linked with numerous health benefits such as cardiovascular protection, antioxidant activity and lowered risk of certain chronic diseases [2,3]. Furthermore, soymilk is low in lactose and cholesterol, making it an ideal choice for people with lactose intolerance, milk allergies and vegetarians and vegans. Given the excellent nutritional, soymilk has become an interesting product in the process of making functional and probiotic fermented foods.

The use of probiotic microorganisms in food systems is a key research field due to their benefits to gut health, immune system and preventing metabolic diseases [4]. Soymilk is an ideal carrier for the probiotic cultures due to its nutritional composition of fermentable carbohydrates, proteins and bioactive compounds which help the growth and metabolism of the microbes. Previous research has shown that probiotic bacteria and bifidobacteria can be successfully cultured in fermented soy products and thus improve the nutritional value and consumer acceptability [3,5].

Additionally, the fermentation of soymilk decreases the beany taste, neutralizes anti-nutrient factors and enhances digestibility, making it more palatable to consume soymilk or food products made from it [5].

However, production of soy yoghurt faces a number of technical difficulties. Soymilk does not contain casein micelles, which are important in the gel formation and development of texture in ordinary yoghurt. As a result, fermented soys tend to have poor body, low viscosity, excessive syneresis and poor sensory properties. Another obstacle to the acceptance of soy-based fermented products is the beany taste of soybeans. Hence, the selection of suitable starter and probiotic cultures that can produce desirable flavour compounds, efficient acidification and enhance textural features is vital to the successful development of soy yoghurt [6].

In the production of fermented products, starter cultures are a crucial step for the quality and functionality. A good starter culture should have good acidification ability and the capacity to produce desirable flavour compounds like acetaldehyde which gives characteristic aroma of yoghurt, and should have rapid growth characteristics [6,7]. Rise in acidification also reduces fermentation time, inhibits undesirable microorganisms and promotes product safety and shelf stability. The growth kinetics such as specific growth rate and generation time, are thus important parameters for the selection of suitable starter cultures for soy yoghurt preparation.

Another important technological characteristic is the production of exopolysaccharides (EPS) by lactic acid bacteria. The extracellular biopolymers, namely EPS, play an important role in the viscosity, water-holding capacity, mouthfeel and structural properties of fermented food products [8]. EPS producing cultures are especially useful in the soy yoghurt production due to their potential of replacing the dairy casein network and optimizing the texture of plant-based fermented products. Some studies have previously shown that the ability of Streptococcus thermophilus and Lactiplantibacillus plantarum to excrete EPS is beneficial for improving rheological properties and consumer acceptability of fermented foods [9].

Technological performance is not the only criterion used in the selection of cultures: probiotic functionality is also a crucial criterion. If the probiotic microorganisms are to have beneficial health effects, they must survive the passage through the gastrointestinal tract and maintain metabolic activity in the gastrointestinal tract. Bile tolerance is thus regarded as one of the most significant attributes for the probiotic potential [10,11]. In addition, cholesterol assimilation has been given a lot of attention due to its cholesterol-lowering potential and its beneficial effects on cardiovascular health [12,13]. Various lactobacilli and bifidobacteria species have been shown to have cholesterol assimilation activity and co-precipitation with bile salts and bile salt hydrolase activity [14].

There is a lack of comparative information of the technological and probiotic properties of the cultures in soy based fermented milk systems, even though several probiotic cultures have been studied in these systems. The selection of cultures for the production of soy yoghurt from the National Collection of Dairy Cultures (NCDC), India has been relatively ignored. The selection of appropriate starter and probiotic cultures for functional soy yoghurt development requires comparative evaluation of the growth kinetics, acidification behaviour, flavour production, EPS synthesis, bile tolerance and cholesterol assimilation of the cultures.

Hence, the present study was conducted to assess and compare the technological and probiotic properties of six different NCDC strains, Lactobacillus acidophilus NCDC15, Lacticaseibacillus casei NCDC17, Lacticaseibacillus rhamnosus NCDC24, Lactiplantibacillus plantarum NCDC25, Bifidobacterium bifidum NCDC232 and Streptococcus thermophilus NCDC293. The specific objectives were to evaluate their growth kinetics, acidification characteristics, production of acetaldehyde, exopolysaccharide production, bile tolerance and cholesterol assimilation, and to determine the most appropriate cultures for making functional fermented soy yoghurt. As far as we know, no previous study has used a Composite Technological–Probiotic Index (CTPI), which combines technological and probiotic properties, for the systematic selection of the NCDC cultures for fermented soy yoghurt production.

2. Materials and Methods

2.1 Microbial Cultures

Six cultures obtained from the National Collection of Dairy Cultures (NCDC), ICAR–National Dairy Research Institute (NDRI), Karnal, India, were used in the investigation. The cultures included Lactobacillus acidophilus NCDC15, Lacticaseibacillus casei NCDC17, Lacticaseibacillus rhamnosus NCDC24, Lactiplantibacillus plantarum NCDC25, Bifidobacterium bifidum NCDC232 and Streptococcus thermophilus NCDC293.

2.2 Activation and Maintenance of Cultures

Freeze-dried cultures obtained from NCDC were activated according to the supplier's recommendations. The lactic cultures were propagated in sterilized de Man, Rogosa and Sharpe (MRS) broth, whereas Streptococcus thermophilus NCDC293 was propagated in sterilized reconstituted skim milk (10%, w/v). The cultures were incubated at 37±1°C for 18–24 h and subsequently transferred twice under the same conditions to obtain active cultures. Activated cultures were maintained at 4°C and subcultured periodically during the experimental period.

2.3 Preparation of Culture Inoculum

Activated cultures were inoculated (2%, v/v) into sterilized soymilk prepared from soybean variety Indira Soya-9 and incubated at 37±1°C. Samples were withdrawn at predetermined intervals for determination of growth characteristics, acidification behaviour and probiotic properties.

2.4 Determination of Growth Characteristics

Growth characteristics were evaluated by monitoring viable cell counts during fermentation. Samples were serially diluted in sterile physiological saline and plated on appropriate growth media. Colony counts were expressed as log CFU mL^{-1}.

The specific growth rate (μ) was calculated during the exponential growth phase using the equation:

$$\mu = \frac{\ln N_2 - \ln N_1}{t_2 - t_1}$$

Where:

N_1 = viable count at time t_1
N_2 = viable count at time t_2
t_1 = initial fermentation time (h)
t_2 = final fermentation time (h)

Generation time (g) was calculated as:

$$g = 0.693/\mu$$

Where:

g = generation time (h)
μ = specific growth rate (h^{-1})

2.5 Determination of Acidification Characteristics

Acidification behaviour of the selected cultures was evaluated during fermentation of sterilized soymilk by monitoring changes in pH and titratable acidity at 0, 3, 6, 9 and 12 h of incubation at 37±1°C. The pH was measured using a calibrated digital pH meter, whereas titratable acidity was determined by titration against 0.1 N sodium hydroxide using phenolphthalein as indicator and expressed as percent lactic acid (% LA).

2.6 Determination of Acetaldehyde Production

Acetaldehyde production was estimated after completion of fermentation using the colorimetric method described by Lees and Jago (1969) [18]. Fermented samples were reacted with appropriate reagents and absorbance was measured spectrophotometrically. Acetaldehyde concentration was calculated from a standard curve and expressed as ppm.

2.7 Determination of Exopolysaccharide (EPS) Production

EPS production was determined following fermentation according to the procedure described by Ruas-Madiedo and de los Reyes-Gavilán (2005). Fermented samples were centrifuged to remove bacterial cells and proteins. EPS present in the supernatant was precipitated using chilled ethanol, recovered by centrifugation and quantified gravimetrically. Results were expressed as mg L^{-1}.

2.8 Determination of Bile Tolerance

Bile tolerance was evaluated by exposing cultures to MRS broth supplemented with 0.3% (w/v) ox-bile. Activated cultures were inoculated into bile-containing medium and incubated at 37±1°C for 4 h. Viable counts were determined before and after incubation.

Bile tolerance (%) was calculated as:

$$\text{Bile tolerance (\%)} = \frac{\text{Viable count after bile exposure}}{\text{Initial viable count}} \times 100$$

2.9 Determination of Cholesterol Assimilation

Cholesterol assimilation was determined using cholesterol-enriched growth medium following the procedure described by Liong and Shah (2005). Activated cultures were inoculated into sterile broth containing soluble cholesterol and incubated at 37±1°C for 24 h. Following incubation, residual cholesterol in the medium was determined spectrophotometrically.

Cholesterol assimilation (%) was calculated as:

$$\text{Cholesterol assimilation (\%)} = \frac{\text{Initial cholesterol} - \text{Residual cholesterol}}{\text{Initial cholesterol}} \times 100$$

2.10 Statistical Analysis

The experiment was conducted in triplicate using a Completely Randomized Design (CRD). Data were analysed by analysis of variance (ANOVA) using OPSTAT statistical software. Treatment means were compared using Critical Difference (CD) at the 5% level of significance. Standard error of mean (SEM±), coefficient of variation (CV%) and significance levels were calculated for all parameters.

3. Results and Discussion

3.1 Screening and Comparative Evaluation of NCDC Cultures

The successful manufacture of fermented soy yoghurt depends largely on the selection of cultures possessing desirable technological and probiotic attributes. Therefore, six cultures obtained from the National Collection of Dairy Cultures (NCDC), namely Lactobacillus acidophilus NCDC15, Lacticaseibacillus casei NCDC17, Lacticaseibacillus rhamnosus NCDC24, Lactiplantibacillus plantarum NCDC25, Bifidobacterium bifidum NCDC232 and Streptococcus thermophilus NCDC293, were comparatively evaluated for their suitability in fermented soy yoghurt manufacture.

The cultures were assessed on the basis of growth kinetics, acidification characteristics, acetaldehyde production, exopolysaccharide (EPS) synthesis and probiotic properties including bile tolerance and cholesterol assimilation. Considerable variation was observed among cultures for all evaluated parameters, indicating significant differences in their fermentation performance and probiotic potential.

Among the cultures evaluated, NCDC293 exhibited the highest growth rate (0.278 h^{-1}), shortest generation time (2.49 h), highest titratable acidity (1.12% LA), maximum acetaldehyde production (30.4 ppm) and greatest EPS production (198.2 mg/L). These characteristics indicate superior fermentation efficiency, flavour development and texture-enhancing potential. Rapid growth and acidification are particularly desirable in fermented soy products because they reduce fermentation time and improve microbial stability [6].

NCDC25 demonstrated excellent technological performance and simultaneously exhibited the superior probiotic characteristics. The culture recorded the highest bile tolerance (94.8%) and cholesterol assimilation (61.2%), indicating superior survival potential under gastrointestinal conditions and enhanced functional properties. Similar observations have been reported for Lactiplantibacillus plantarum, which is recognized for its strong probiotic functionality and metabolic versatility [11,15].

NCDC24 also exhibited favourable technological and probiotic characteristics, ranking intermediate between NCDC25 and NCDC293. In contrast, NCDC232 recorded the lowest performance for most parameters, including growth rate, acetaldehyde production, EPS synthesis, bile tolerance and cholesterol assimilation.

TABLE 1: TECHNOLOGICAL AND PROBIOTIC CHARACTERISTICS OF SIX NCDC CULTURES EVALUATED FOR FERMENTED SOY YOGHURT MANUFACTURE

Culture	*Growth Rate (h^{-1})*	*Generation Time (h)*	*Titratable Acidity (% LA)*	*Acetaldehyde (ppm)*	*EPS (mg L^{-1})*	*Bile Tolerance (%)*	*Cholesterol Assimilation (%)*
NCDC15 (*Lactobacillus acidophilus*)	0.229	3.03	0.84	18.4	118.4	86.4	42.6
NCDC17 (*Lacticaseibacillus casei*)	0.245	2.83	0.95	21.2	136.7	89.2	48.4
NCDC24 (*Lacticaseibacillus rhamnosus*)	0.255	2.72	0.98	23.5	152.3	91.6	53.8
NCDC25 (*Lactiplantibacillus plantarum*)	0.268	2.59	1.06	27.1	184.6	94.8	61.2
NCDC232 (*Bifidobacterium bifidum*)	0.205	3.38	0.78	14.2	104.8	82.7	38.9
NCDC293 (*Streptococcus thermophilus*)	0.278	2.49	1.12	30.4	198.2	87.5	45.7

Parameter	*SEM±*	*CD (P≤0.05)*	*CV (%)*
Growth Rate (h^{-1})	0.003	0.011	1.777
Generation Time (h)	0.021	0.066	1.784
Titratable Acidity (% LA)	0.006	0.018	1.040
Acetaldehyde (ppm)	0.388	1.209	2.991
EPS (mg L^{-1})	2.902	9.042	3.370
Bile Tolerance (%)	1.090	3.396	2.129
Cholesterol Assimilation (%)	0.854	2.662	3.055

Note: Values represent means of three replications. Titratable acidity is expressed as percentage lactic acid (% LA). EPS = exopolysaccharide production. Growth rate and generation time were calculated from exponential growth kinetics. Statistical analysis was performed using a Completely Randomized Design (CRD). SEM± = standard error of mean; CD = critical difference; CV = coefficient of variation. Means differing by more than the corresponding CD value are significantly different at P≤0.05.

3.2 Selection of Cultures for Soy Yoghurt Manufacture

The data presented in Table 1 were subsequently normalized and used for calculation of the Composite Technological–Probiotic Index (CTPI).

To facilitate objective comparison among cultures, all parameters were normalized using min–max scaling (Table 2). Beneficial parameters were normalized according to:

For beneficial parameters

$$\text{Normalized Score} = \frac{X - X_{min}}{X_{max} - X_{min}}$$

For generation time

†Generation time was reverse-normalized because lower values indicate superior performance:

$$\text{Normalized Score} = \frac{X_{max} - X}{X_{max} - X_{min}}$$

TABLE 2: NORMALIZED SCORES OF NCDC CULTURES

Culture	*Growth Rate Score*	*Generation Time Score†*	*TA Score*	*Acetaldehyde Score*	*EPS Score*	*Bile Tolerance Score*	*Cholesterol Assimilation Score*
NCDC15 (*L. acidophilus*)	0.329	0.411	0.176	0.272	0.139	0.296	0.155
NCDC17 (*L. casei*)	0.548	0.616	0.500	0.471	0.337	0.533	0.444
NCDC24 (*L. rhamnosus*)	0.685	0.726	0.588	0.635	0.522	0.737	0.743
NCDC25 (*L. plantarum*)	0.863	0.863	0.824	0.888	0.847	1.000	1.000
NCDC232 (*B. bifidum*)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NCDC293 (*S. thermophilus*)	1.000	1.000	1.000	1.000	1.000	0.407	0.339

Note: Parameters (growth rate, titratable acidity, acetaldehyde production, EPS production, bile tolerance and cholesterol assimilation) were normalized using min–max scaling. Generation time was reverse-normalized because lower values indicate superior performance. Composite Technological–Probiotic Index (CTPI) was calculated as the arithmetic mean of seven normalized scores according to the equation presented in Section 3.2.

Composite Technological–Probiotic Index (CTPI)

Equal weighting was assigned to all seven parameters because each characteristic contributes substantially to the technological and probiotic suitability of cultures for fermented soy yoghurt manufacture.

$$\text{CTPI}=\frac{\sum_{i=1}^{n} S_i}{n}$$

Where:

X = observed value of the parameter
X_{min} = minimum value among all cultures
X_{max} = maximum value among all cultures
Si = normalized score of the ith parameter

n = 7 parameters

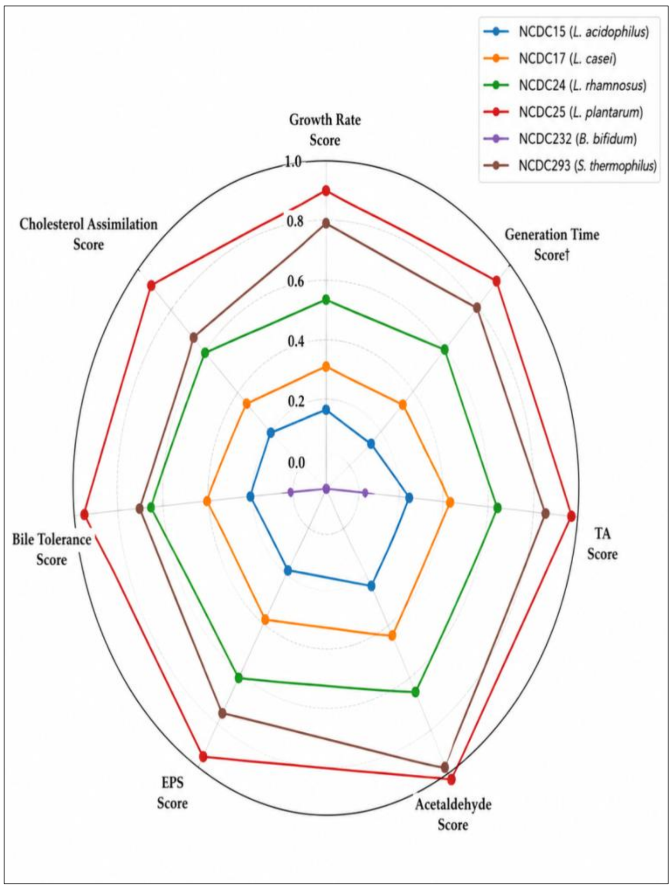

FIG. 1: RADAR CHART SHOWING NORMALIZED TECHNOLOGICAL AND PROBIOTIC SCORES OF SIX NCDC CULTURES

Figure 1 illustrates the normalized technological and probiotic performance of the six NCDC cultures. NCDC25 and NCDC293 occupied the largest radar area, indicating superior overall performance compared with the remaining cultures. NCDC25 exhibited the highest probiotic functionality, particularly in bile tolerance and cholesterol assimilation, whereas NCDC293 demonstrated superior technological characteristics including growth rate, acidification capacity, acetaldehyde production and EPS synthesis. NCDC24 showed balanced technological and probiotic performance and ranked third overall. In contrast, NCDC232 exhibited the lowest composite performance across most evaluated parameters. These observations supported the selection of NCDC25, NCDC293 and NCDC24 for subsequent evaluation in soymilk fermentation (Table 3).

TABLE 3: COMPOSITE TECHNOLOGICAL–PROBIOTIC INDEX (CTPI), RANKING AND SELECTION STATUS OF NCDC CULTURES

Culture	*CTPI*	*Rank*	*Selection Status*
NCDC25	0.884	1	Selected
NCDC293	0.821	2	Selected
NCDC24	0.662	3	Selected
NCDC17	0.493	4	Not Selected
NCDC15	0.254	5	Not Selected
NCDC232	0.000	6	Not Selected

Note: CTPI values were calculated as the arithmetic mean of seven normalized scores representing growth rate, generation time, titratable acidity, acetaldehyde production, EPS production, bile tolerance and cholesterol assimilation. Higher CTPI values indicate greater suitability for fermented soy yoghurt manufacture.

The normalized scoring approach provided an integrated assessment of technological and probiotic performance among the six NCDC cultures. NCDC25 achieved the highest CTPI (0.884), followed by NCDC293 (0.821) and NCDC24 (0.662). These cultures consistently exhibited superior performance across multiple technological and probiotic attributes. NCDC293 demonstrated exceptional technological performance characterized by rapid growth, strong acidification, elevated acetaldehyde production and high EPS synthesis, whereas NCDC25 exhibited superior probiotic functionality with the highest bile tolerance and cholesterol assimilation. NCDC24 demonstrated balanced technological and probiotic characteristics and ranked third overall.

Based on CTPI values and overall ranking, Lactiplantibacillus plantarum NCDC25, Streptococcus thermophilus NCDC293 and Lacticaseibacillus rhamnosus NCDC24 were selected for subsequent evaluation in soymilk fermentation. These cultures represented the most desirable combination of technological efficiency and probiotic functionality among the six cultures investigated.

3.3 Evaluation of Selected Cultures in Soymilk

The three highest-ranked cultures identified during the screening phase, namely Streptococcus thermophilus NCDC293, Lactiplantibacillus plantarum NCDC25 and Lacticaseibacillus rhamnosus NCDC24, were further evaluated in standardized soymilk to assess their fermentation performance under actual product conditions. Growth characteristics, acidification behaviour, acetaldehyde production and exopolysaccharide (EPS) synthesis were determined and the results are presented in Table 4 and Figures 2-3.

All three cultures adapted successfully to the soy substrate and exhibited substantial increases in viable cell populations during fermentation.

TABLE 4: GROWTH, ACIDIFICATION AND METABOLITE PRODUCTION CHARACTERISTICS OF SELECTED CULTURES IN SOYMILK

Parameter	*NCDC293*	*NCDC25*	*NCDC24*
Final viable count (log CFU/mL)	9.28	9.00	8.83
Growth rate (μ h^{-1})	0.312	0.273	0.248
Generation time (h)	2.22	2.54	2.79
Final pH	4.45	4.65	4.85
Titratable acidity (% LA)	0.820	0.720	0.640
Acetaldehyde (ppm)	26.40	21.30	16.77
EPS (mg L^{-1})	198.80	165.70	134.00
Parameter	***SEM±***	***CD (P≤0.05)***	***CV (%)***
Final viable count (log CFU/mL)	0.011	0.038	0.205
Growth rate (μ h^{-1})	0.001	0.004	0.721
Generation time (h)	0.006	0.020	0.398
Final pH	0.012	0.041	0.430
Titratable acidity (% LA)	0.006	0.020	1.376
Acetaldehyde (ppm)	0.150	0.530	1.212
EPS (mg L^{-1})	0.347	1.223	0.361

Note: Values represent treatment means obtained after 12 h fermentation of standardized soymilk. Significant differences among cultures were observed for all parameters (P≤0.05). Viable counts are expressed as log CFU/mL, titratable acidity as % lactic acid, acetaldehyde as ppm and EPS as mg L^{-1}.

Analysis of variance revealed significant (P≤0.05) differences among cultures for all evaluated parameters, confirming strain-dependent variation in growth, acidification, flavour development and EPS production in soymilk.

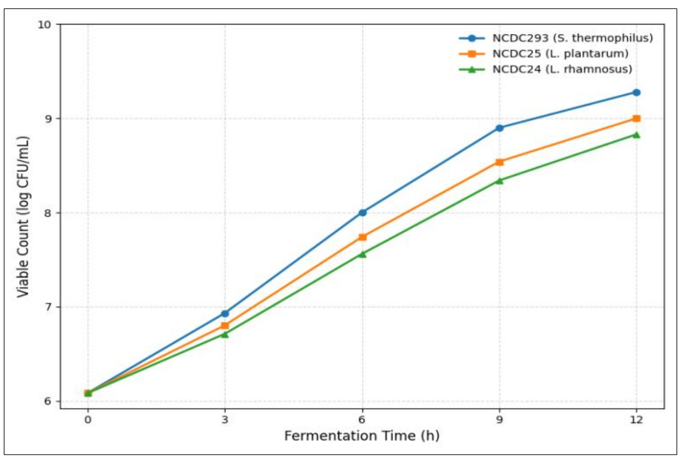

FIG. 2: GROWTH CURVES OF SELECTED CULTURES DURING SOYMILK FERMENTATION

All three cultures adapted successfully to the soy substrate and exhibited substantial increases in viable cell populations during fermentation. Initial counts of approximately 6.08 log CFU/mL increased to 9.28, 9.00 and 8.83 log CFU/mL for NCDC293, NCDC25 and NCDC24, respectively, after 12 h fermentation. NCDC293 showed significantly higher growth than the probiotic cultures throughout fermentation, indicating superior adaptation to the soymilk environment. Similar growth behaviour of starter and probiotic cultures in fermented soy beverages has been reported previously [16,17].

Growth kinetic analysis further confirmed the superior performance of NCDC293. The culture exhibited the highest specific growth rate (0.312 h^{-1}) and shortest generation time (2.22 h), followed by NCDC25 (0.273 h^{-1}; 2.54 h) and NCDC24 (0.248 h^{-1}; 2.79 h). Faster growth kinetics facilitate rapid fermentation and improved process efficiency. The higher viable count and growth rate of NCDC293 observed in soymilk were consistent with its superior technological performance during the initial culture screening stage.

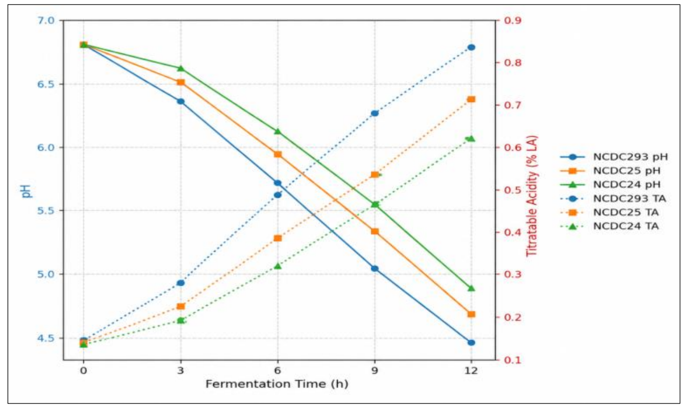

FIG. 3: ACIDIFICATION PROFILES OF SELECTED CULTURES DURING SOYMILK FERMENTATION SHOWING CHANGES IN PH AND TITRATABLE ACIDITY (% LACTIC ACID) OVER 12 h INCUBATION

Acidification behaviour differed significantly among cultures. NCDC293 reduced the pH of soymilk from 6.80 to 4.45 and simultaneously increased titratable acidity to 0.820% lactic acid after 12 h fermentation. Corresponding values for NCDC25 and NCDC24 were 4.65 and 4.85 pH units, with titratable acidity values of 0.720 and 0.640%, respectively. Rapid acidification is desirable because it promotes protein coagulation, enhances microbial safety and improves product stability [6].

Significant differences were also observed in flavour and texture-related metabolites. NCDC293 produced the highest concentration of acetaldehyde (26.40 ppm), followed by NCDC25 (21.30 ppm) and NCDC24 (16.77 ppm). Acetaldehyde is the principal aroma compound responsible for the characteristic yoghurt flavour and contributes to masking undesirable beany notes in fermented soy products. Similarly, EPS production was highest in NCDC293 (198.80 mg L^{-1}), followed by NCDC25 (165.70 mg L^{-1}) and NCDC24 (134.00 mg L^{-1}). Elevated EPS production is advantageous because it improves viscosity, water-holding capacity and texture of fermented soy yoghurt.

Overall, NCDC293 demonstrated superior technological performance in soymilk, characterized by rapid growth, efficient acidification, high acetaldehyde production and elevated EPS synthesis. NCDC25 and NCDC24 exhibited satisfactory growth and fermentation performance while retaining the superior probiotic characteristics identified during the screening stage. Therefore, NCDC293 was selected as the principal starter culture, whereas NCDC25 and NCDC24 were selected as adjunct probiotic cultures for subsequent soy yoghurt manufacture. The complementary technological and probiotic characteristics of these cultures justified their use as a mixed-culture system for subsequent soy yoghurt development and optimization studies.

The present investigation evaluated individual cultures under laboratory conditions. Further studies are required to assess mixed-culture interactions, sensory acceptability, storage stability and probiotic survival during refrigerated storage.

CONCLUSION

There was a huge variance in the technological and probiotic properties among the six NCDC cultures in this comparative evaluation. Streptococcus thermophilus NCDC293 showed better technological properties, such as high growth rate, high acidification, production of high levels of acetaldehyde, and high EPS production rate. Lactiplantibacillus plantarum NCDC25 was found to possess the best probiotic potential because of its highest bile tolerance and cholesterol assimilation.

The most suitable cultures for fermented soy yoghurt manufacture were determined to be NCDC25, NCDC293 and NCDC24 from the Composite Technological–Probiotic Index (CTPI). NCDC293 showed a high technological performance while NCDC25 showed the highest probiotic potential. Synergistic effect of these cultures can enhance the fermentation efficiency, functional and quality properties of fermented soy yoghurt.

The lacticaseibacillus rhamnosus NCDC24 strain showed good technology and probiotic properties, and it can be used as an appropriate adjunct culture in the mixed culture fermented soy yoghurt systems.

ACKNOWLEDGEMENT

The authors acknowledge the National Collection of Dairy Cultures (NCDC), ICAR-NDRI, Karnal, India, for providing microbial cultures used in the investigation.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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How to cite this article:
Kumar M, Kumar S, Manish. Comparative evaluation and selection of ncdc cultures for fermented soy yoghurt manufacture based on technological and probiotic characteristics. P-Edu International Journal of Multidisciplinary Studies 2026; 2(1): 37-43.