Hindawi
Journal of Food Quality
Volume 2023, Article ID 6687015, 12 pages
https://doi.org/10.1155/2023/6687015

*Research Article*

# Characterization of Lactic Acid Bacteria and Pathogens Isolated from Traditionally Fermented Foods, In Relation to Food Safety and Antimicrobial Resistance in Tribal Hill Areas of Northeast India

**Tushar Kumar Dey**,[1,2,3] **Johanna F. Lindahl**,[1,2,4] **Rajkumari Sanjukta**,[3] **A. Arun Prince Milton**,[3] **Samir Das**,[3] **Porteen Kannan**,[5] **Åke Lundkvist**,[2] **Arnab Sen**,[3] **and Sandeep Ghatak**[3]

[1]*Department of Biosciences, International Livestock Research Institute, Nairobi, Kenya*
[2]*Department of Medical Biochemistry and Microbiology, Zoonosis Science Center, Uppsala University, Uppsala, Sweden*
[3]*Division of Animal and Fisheries Sciences, ICAR Research Complex for North-eastern Hill Region, Ri-Bhoi, Meghalaya, India*
[4]*Department of Clinical Sciences, Swedish University of Agricultural Sciences, Uppsala, Sweden*
[5]*Department of Veterinary Public Health and Epidemiology, Madras Veterinary College, TANUVAS, Chennai, India*

Correspondence should be addressed to Sandeep Ghatak; ghataksnd@rediffmail.com

Received 20 January 2023; Revised 11 April 2023; Accepted 9 May 2023; Published 26 May 2023

Academic Editor: Ali Akbar

Copyright © 2023 Tushar Kumar Dey et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Traditional fermented food products are often connected to various indigenous tribes and thus vary due to ethnicity, geography, and natural resource availability. The indigenous tribes from India greatly rely on fermentation processes for food preservation, flavor, and nutrition. Fermented foods can provide health benefits but also pose risks from harmful microbes and contaminants that grow in the food due to poor hygiene. In this study, we identified lactic acid bacteria (LAB) in fermented food collected from Northeast India, assessed their beneficial properties, and highlighted the risk from food pathogens that have antimicrobial resistance traits. A total of 113 different samples of fermented food products were collected from the local markets of five Northeastern Indian states (Nagaland, Manipur, Meghalaya, Arunachal Pradesh, and Sikkim). Standard laboratory methods were used to isolate LAB and determine their probiotic properties, conduct coliform counts, and isolate presumptive staphylococci from the fermented food samples. Antimicrobial susceptibility was determined by using the BD-Phoenix 100 automated system. We isolated 30 LAB with probiotic potential. The average aerobic colony count in different fermented food was 4.4–7.7 log·cfu/g, while coliforms were present in 43% (49/113, 95% (CI 34–53)) of the food samples, indicating low-hygiene levels. Additionally, some food samples contained staphylococci with phenotypic antibiotic-resistance markers (MRS, HLMUP, BLACT, and STAMLS). This study indicates that probiotic bacteria could be present in traditional fermented food products of Northeast India, but contamination with staphylococci and other bacterial pathogens with antibiotic resistance traits could put the health of consumers at risk.

## 1. Introduction

Historically, people have used various methods to preserve and enhance the shelf life of food; one extensively used practice is fermentation [1]. Fermentation involves the activity of microorganisms which play a vital role in the elimination of many pathogens and help preserve food products [2]. Some lactic acid bacteria (LAB) involved in fermentation are probiotic in nature and beneficial for human health when consumed in adequate quantities via food products [3]. LAB enhances the nutritional and therapeutic properties of the food products to provide

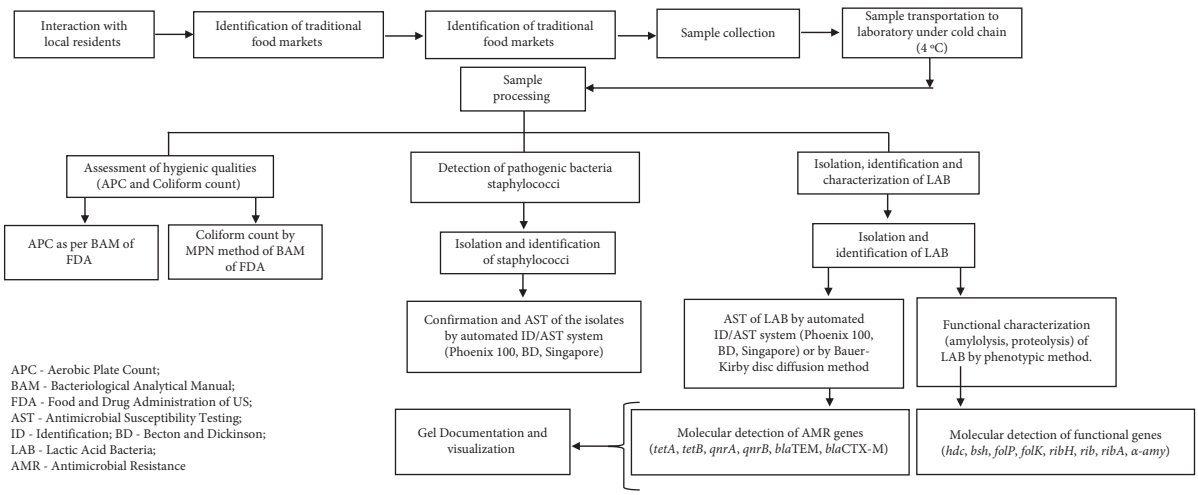


Figure 1: Flowchart for isolation of lactic acid bacteria (LAB), hygiene quality determination, and molecular characterization of isolates from fermented food products.

beneficial health effects and play a vital role as gut microbiota [4, 5]. Fermentation helps improve food digestion by breaking down complex sugars into simpler forms, as well as providing essential amino acids, vitamins, and minerals, and can improve the flavor and taste [1, 2]. Protein and vitamin deficiencies are major problems in low- and middle-income countries [6, 7], and these populations could potentially be helped by increased consumption of fermented products that can help meet the requirement for healthy gut microbiota and mitigate nutrition deficiencies [8–12]. Traditional fermented food products are often connected to various indigenous tribes [13]. Countries like India have many tribal populations, and these tribes represent varieties of traditional fermented food due to varied ethnicity, geography, and natural resource availability [14]. Fermented foods have played a vital role in contributing to Indian diets over many centuries, partly because fermentation is a cheap and cost-effective food preservation technique [15]. The tribal people of Indian states use ethnic traditional knowledge inherited from their ancestors to prepare fermented food [16]. Although fermented foods can provide health benefits [1, 17], there is also a risk of harmful microorganisms that may grow in the food products [18] due to poor hygiene during food preparation or storage and cause adverse effects to human health [19]. Fermentation should be studied and developed further so that fermented products can be of standardized quality and safer for consumers [20–23]. Reconsidering the food safety aspects of fermented food preparation would be beneficial to producers and consumers, and this could generate a more sustainable source of income for communities. However, there is a paucity of data on the food safety aspects of the fermented foods that are prepared and consumed by various tribes of Northeast India. Therefore, the present study was undertaken to assess various food safety parameters in fermented food products and the beneficial effects of the isolated LAB and identify associated pathogens, specifically, staphylococci, while also investigating their antimicrobial resistance traits.

## 2. Materials and Methods

The samples of fermented food products were collected and processed for isolation of indigenous LAB and associated food pathogens, followed by their identification. LAB was studied for their functional and probiotic properties by phenotypic and genotyping methods as described in the flowchart (Figure 1).

*2.1. Sampling.* One hundred and thirteen samples of fermented food products were collected from five Northeastern Indian states (Nagaland, Manipur, Meghalaya, Arunachal Pradesh, and Sikkim) (Table 1). The samples were collected from traditional markets using a nonprobabilistic sampling method (convenience sampling) over a two-year period (2013 to 2015). The most prominent traditional food markets in each of the five states were identified through interactions with local residents. The fermented food samples were chosen by the vendor from a basket made of bamboo, and the samples were sometimes found wrapped in a leaf. The collected samples were placed in sterile 250 ml zip lock pouches, which were transported to the laboratory under refrigeration temperature (4°C). On arrival at the laboratory, the samples were immediately processed for further microbiological analyses. The interval between the collection of samples and the onset of analysis did not exceed 48 hours. Each sample was aseptically divided into two parts. One part was processed for isolation of LAB, and the other was analyzed for aerobic plate counts and coliform counts to assess the hygienic status.

*2.2. Microbiological Analyses*

*2.2.1. Isolation of LAB.* For isolation of LAB, the procedures reported by Tamang et al. [24] were followed with minor adaptations. Following aseptic procedures, 25 g of each sample was weighed and homogenized in 225 ml of sterile normal saline solution (0.85% NaCl) in a conical flask. The

Table 1: Details of the fermented food type, number of samples, and local name in brackets, from the five different states.

| Fermented food | Nagaland | Arunachal Pradesh | Sikkim | Meghalaya | Manipur | Total |
|---|---|---|---|---|---|---|
| Dry fish | 9 (Sukamas) | 0 | 0 | 5 (Tungtap) | 15 (Ngari) | 29 |
| Bamboo shoot | 35 (Banstenga) | 2 (Ikung) | 0 | 0 | 6 (Soibum) | 43 |
| Soyabean | 30 (Axone) | 2 (Peruyyan) | 0 | 0 | 4 (Hawaijar) | 36 |
| Fish pickle | 0 | 1 (Ngakam Azey) | 0 | 0 | 0 | 1 |
| Meat pickle | 0 | 1 (Shakam Azey) | 0 | 0 | 0 | 1 |
| Yak cheese | 0 | 1 (Chhurpi) | 0 | 0 | 0 | 1 |
| Milk | 0 | 0 | 2 (Chhu) | 0 | 0 | 2 |
| Total | 74 | 7 | 2 | 5 | 25 | 113 |

flask was left undisturbed for 10 minutes at room temperature for settling of coarse particles. From the homogenate, 10 ml was inoculated in 90 ml of sterile De Man, Rogosa, and Sharpe (MRS) broth (Hi-media, Maharashtra, India) and incubated at 37°C for 18–24 hours. Following incubation, 1 ml of enriched broth was inoculated in duplicate on MRS agar plates supplemented with 1% $CaCO_3$. Of the duplicate MRS agar plates for each sample, one set was incubated under anaerobic conditions (Gas Pack-LE002A, Hi-media, Maharashtra, India) in an anaerobic jar at 30°C for up to 3 days. The other set of agar plates was incubated aerobically at 37°C for 24–48 hours. Presumptive colonies of LAB were identified by colony morphology (white colony with a clearing zone of $CaCO_3$ deposition in MRS agar) and further tests including Gram staining, catalase, and oxidase production as per standard protocols [25]. A purity check of the isolates was performed by subculturing the presumptive isolates on fresh MRS agar plates, followed by microscopic examinations (Leica, Wetzlar, Germany). Presumptive isolates were then identified using the Phoenix 100 automated microbiological identification system (Becton and Dickinson, Tuas, Singapore).

*2.2.2. Assessment of Hygienic Quality by the Aerobic Plate Count (APC) and Most Probable Number (MPN) of Coliforms.* To assess the hygienic status of the fermented food samples, APC was determined following the procedure described in the Bacteriological Analysis Manual of Food and Drug Administration [26]. Each sample was tested on duplicate plates of plate count agar (Hi-media, Maharashtra, India), and the average count was recorded for interpretation. In addition to the aerobic plate count (APC), coliform counts were determined by the most probable number (MPN) as described previously for food samples [27]. Each sample was processed using three tube tests (1, 0.1 and 0.01 ml), and the MPN indices were recorded. The samples which were negative for all the three tubes had a MPN value < 0.3 and were assigned 0.15 in the quantitative calculation.

*2.2.3. Isolation and Identification of Pathogens.* Isolation of pathogens from fermented foods was undertaken by the procedures described previously with suitable modifications [28]. The isolates that grew along with LAB and were identified as staphylococci were further reconfirmed by inoculating in Brain Heart Infusion (BHI) agar (Hi-media, Maharashtra, India), followed by overnight incubation at 37°C. The bacterial colonies from BHI agar were streaked on Baird Parker Agar plates (Hi-media, Maharashtra, India) and reincubated at 37°C for 18 hours. Presumptive colonies of staphylococci (grey-black to black, with or without shine and halo) were picked (one random colony for each sample). Other suspected colonies from BHI agar were also randomly picked and characterized by using Phoenix 100 automated ID/AST system (Becton and Dickinson, Tuas, Singapore) for confirmatory identification.

### 2.3. Functional Characterization of LAB by Phenotypic Tests

*2.3.1. Test for Proteolysis.* Isolates of LAB were inoculated in MRS broth and incubated for 24 hours at 30°C. Following incubation, bacteria were harvested by centrifugation ($8000 \times g$). Resulting pellets were washed three times in sterile phosphate buffered saline (1X PBS) (pH 7.2) and resuspended in 1 ml of PBS. The optical density of the suspension was adjusted to 0.5 MacFarland using a nephelometer (BD PhoenixSpec). Ten microliters of bacterial suspension were inoculated into each well (6 mm diameter) made in the skim milk agar (Hi-media, Maharashtra, India) plates. The plates were incubated at 37°C for 4 h and analyzed for the presence or absence of a clear zone of proteolysis around the inoculated wells. For each plate, a positive control (*Pseudomonas aeruginosa* ATCC 27853) and a negative control (*Escherichia coli* ATCC 25922) were included [29].

*2.3.2. Test for Amylolysis.* The amylolytic activity of the LAB isolates was estimated with bacterial suspension prepared as described in Section 2.3.1. Surface-dried plates of starch agar (Hi-media, Maharashtra, India) were inoculated with 10 $\mu$l of bacterial suspension and incubated at 37°C for 48 h. After incubation, a few drops of Gram's iodine solution (Hi-media, Maharashtra, India) were added onto the agar surface, and the plate was allowed to stand for 10 minutes. Positive results were noted as clear zones of starch hydrolysis around the inoculation sites. In each experiment, suspensions of *Aeromonas hydrophila* ATCC 7966 and *E. coli* ATCC 25922 were included as positive and negative controls, respectively.

*2.4. Antimicrobial Susceptibility Test.* Susceptibility of LAB and identified pathogenic bacteria to antibiotics was tested as described elsewhere [30–32]. Following presumptive

Table 2: Details of PCR primers for identification and detection of functional and AMR genes.

| General functions | Gene | Primer | | *AT (°C) | References |
|---|---|---|---|---|---|
| | | Orientation | Sequence 5′ to 3′ | | |
| Survival pH | *hdc* | F | AGATGGTATTGTTTCTTATG | 52 | [34] |
| | | R | AGACCATACACCATAACCTT | | |
| Bile salt | *Bsh* | F | ATTGAAGGCGGAACSGGMTA | 58 | |
| | | R | ATWACCGGWCGGAAAGCTG | | |
| Synthesis of B vitamins | *folP* | F | CCASGRCSGCTTGCATGAC | 59.5 | |
| | | R | TKACGCCGGACTCCTTTTWY | | |
| | *folK* | F | CCATTTCCAGGTGGGGAATC | 59.5 | |
| | | R | GGGGTGGTCCAAGCAAACTT | | |
| | *ribH* | F | AGGGCGAAACCGACCACTAC | 60 | |
| | | R | CGATTGGGCAGTCATCGAAC | | |
| Riboflavin synthesis | *ribB* | F | AGTAAACGGAACGGGCAAGC | 60 | |
| | | R | GTTGACCAGGGCACCAACTG | | |
| | *ribA* | F | TTTACGGGCGATGTTTTAGG | 60 | |
| | | R | CGACCCTCTTGCCGTAAATA | | |
| Starch metabolism | *α-amy* | F | AGATCAGGCGCAAGTTCAGT | 60 | |
| | | R | TTTTATGGGCACACCACTCA | | |
| Tetracycline | *tetA* | F | GTAATTCTGAGCACTGTCGC | 57 | [35] |
| | | R | CTGTCCTGGACAACATTGCTT | | |
| | *tetB* | F | CTCAGTATTCCAAGCCTTTG | 52 | |
| | | R | CTAAGCACTTGTCTCCTGTT | | |
| Quinolone | *qnrA* | F | GGATGCCAGTTTCGAGGA | 59 | [36] |
| | | R | TGCCAGGCACAGATCTTG | | |
| | *qnrB* | F | GGMATHGAAATTCGCCACTG | 55 | [37] |
| | | R | TTTGCYGYYCGCCAGTCGAA | | |
| *β*-Lactamase | *blaTEM* | F | ATGAGTATTCAACATTTCCG | 55 | [38] |
| | | R | TTAATCAGTGAGGCACCTAT | | |
| | *CTX-M* | F | CGCTTTGCGATGTGCAG | 60 | |
| | | R | ACCGCGATATCGTTGGT | | |

*AT; annealing temperature.

identification, isolates were characterized by using the BD Phoenix-TM 100 automated ID/AST system (Becton and Dickinson, Tuas Avenue, Singapore) for minimum inhibitory concentrations of various antimicrobials employing Gram-positive combo panels (PMIC/ID-55) following the Clinical and Laboratory Standards Institute (CLSI) guidelines [33]. Manual antibiotic susceptibility testing (AST) was performed as per the Bauer–Kirby disc-diffusion test for the bacterial isolates that were not included in BD Phoenix AST taxonomy. This was performed by inoculating the overnight grown cultures on Muller–Hinton agar (MHA) (Hi-media, Maharashtra, India) plates. The bacterial suspension was set to 0.5 McFarland prior to inoculation on MHA agar. A sterile swab was used to uniformly spread the culture on the agar plate. Post inoculation, the antibiotic discs (Hi-media, Maharashtra, India) with specific antibiotic concentrations were placed on the surface of the agar plates with sterile forceps and incubated at 37°C for 18–24 hours. Isolates were considered as multidrug resistant if they show resistance to more than three classes of antibiotics or if resistance markers such as BLACT (beta-lactamase), MRS (methicillin-resistant staphylococci), HLGR (high-level gentamicin resistance), HLMUP (high-level mupirocin), mecA-RS (mecA-mediated resistant S*taphylococcus*), STAMLS (*Staphylococcus* MLSb: macrolides, lincosamides, and streptogramin B) and VRE (vancomycin resistant enterococci) carbapenems and nitrofurantoin resistance were indicated in AST results by using the automated BD Phoenix system.

### 2.5. Molecular Characterization

*2.5.1. Isolation of Genomic DNA.* Bacterial genomic DNA was extracted using a column-based bacterial genomic DNA extraction kit (Hi-media, Maharashtra, India) according to the manufacturer's instructions.

*2.5.2. Polymerase Chain Reaction.* A ThermoFisher Scientific (Waltham, Massachusetts, USA) thermal cycler was used for all polymerase chain reaction (PCR) analyses. PCR was carried out to detect the presence of the following genes, for riboflavin production (*ribH, ribB,* and *ribA* genes), for tolerance to bile salt (*bsh* gene), for starch metabolism (*amy* gene), for synthesis of B vitamins, including folate synthesis (*folP and folk* genes), and for riboflavin synthesis (*rib, ribH,* and *ribA* genes) [34] by specific primers (Table 2). The AMR genes (*tetA*, *tetB*, *tetC*, *TEM, CTX-M*, *vanA*, and *vanB* genes) were screened by using the primers listed in Table 2.

## 3. Results

*3.1. Identification of LAB.* LAB were isolated from 20 of the 113 (17.7%; 95% confidence interval (CI) (11–26%)) samples. In total, 30 LAB (including duplicates) were identified by using the BD PhoenixTM100 automated ID/AST system as *Pediococcus parvulus* (10), *Pediococcus pentosaceus* (9), *Pediococcus acidilactici* (5), *Leuconostoc mesenteroides* (3), *Pediococcus damnosus* (1), *Aerococcus viridans* (1), and *Lactococcus plantarum* (1). The dominant genus in the fermented products was *Pediococcus* (isolated from 9 samples of fermented dry fish and 8 samples of fermented bamboo shoots). No LAB were isolated from yak cheese, milk products, and fish and meat pickles.

*3.2. APC and MPN of Coliforms.* The fermented food samples showed varied aerobic colony counts. The average APC was found to be highest in fermented soyabean (average 7.7 log cfu/g), followed by fermented milk (average 6.4 log cfu/g), meat pickles (average 6.1 log cfu/g), yak cheese (average 5.9 log cfu/g), fish pickles (average 5.1 log cfu/g), and dry fish (average log 5.1 cfu/g). The lowest colony count was observed in fermented bamboo shoots (average 4.4 log cfu/g) (Table 3), (Supplementary Materials, Table S1). According to the compendium of microbiological criteria for food 2018 [39], fermented food falls under category 5, and there are no interpretation criteria available to interpret the result as satisfactory, marginal, or unsatisfactory.

The overall coliform prevalence in the fermented food samples was 43% (49/113; 95% CI (34–53)). Coliforms were detected in all tested samples of fermented milk, fish pickle, and meat pickle, while the prevalence levels of coliforms were higher in soyabeans (67%), followed by dry fish (39%) and bamboo shoots (22%). The average concentration of coliforms in the fermented food samples was 0.50 log MPN/ml (g), and the range was from −0.82 to 0.04 log MPN/ml (g) (Table 4) (Supplementary Materials, Table S1).

*3.3. Identification of Pathogens.* Fifty-three percent (60/113; 95% CI (42–63)) of the fermented food samples were found to contain different pathogens (Table 5). A total of 145 presumptive pathogens (including duplicates) were found in the samples identified by using the BD Phoenix ID/AST system. Among the isolated presumptive pathogens, only staphylococci were reconfirmed by growing them in a selective medium followed by microscopic examination and reidentified using the BD phoenix system. The rest of the isolates were not reconfirmed and hence cannot be concluded to be pathogens (Table 5), (Supplementary Materials, Table S1).

*3.4. Proteolytic and Amylolytic Activity by the Phenotypic Method.* Amylolytic activity was observed in 33% (10/30; 95% (CI 17–53)) of LAB, and none of the LAB isolates exhibited proteolytic activity. Amylolytic activity was observed in the following LAB: *Pediococcus pentosaceus* (5), *Pediococcus acidilactici* (1), *Leuconostoc mesenteroides* (2), *Lactococcus plantarum* (1), and *Pediococcus parvulus* (1) (Supplementary Materials, Table S2).

*3.5. Molecular Detection of Functional and Antibiotic Resistance Genes by PCR.* By screening the LAB (30) for the functional genes, 80% (24/30; 95% CI (50–85)) possessed riboflavin synthesis capacity (*ribA* gene), 20% (6/30; 95% CI (7–38)) had the combination (*ribA* + *ribB* genes), 70% (21/

Table 3: Aerobic plate counts in fermented food samples, presented as the average, minimum, and maximum of the log colony-forming units (cfus).

| Fermented food | Samples | Average of log cfu/g | Minimum of log cfu/g | Maximum of log cfu |
|---|---|---|---|---|
| Dry fish | 31 | 5.11 | 1.00 | 10.48 |
| Bamboo shoots | 41 | 4.43 | 1.00 | 9.03 |
| Soyabeans | 36 | 7.68 | 1.00 | 10.05 |
| Fish pickles | 1 | 5.12 | 5.12 | 5.12 |
| Meat pickles | 1 | 6.05 | 6.05 | 6.05 |
| Milk products | 2 | 6.50 | 6.35 | 6.64 |
| Yak cheese | 1 | 5.87 | 5.87 | 5.87 |
| Overall | 113 | 5.72 | 1.00 | 10.48 |

Table 4: Prevalence and concentration (log most probable number (MPN)/ml (g)) of coliforms in tested fermented food samples.

| Fermented food | Total samples | Positive sample | Coliform prevalence (%) | Concentration (mean (SD), log MPN/ml (g)) |
|---|---|---|---|---|
| Bamboo shoots | 41 | 9 | 22.0 | −0.67 (0.33) |
| Dry fish | 31 | 12 | 38.70 | −0.55 (0.40) |
| Milk | 2 | 2 | 100.0 | −0.04 (0.12) |
| Soyabeans | 36 | 24 | 66.7 | −0.30 (0.44) |
| Fish pickles | 1 | 1 | 100.0 | 0.04 (NA) |
| Meat pickles | 1 | 1 | 100.0 | 0.04 (NA) |
| Yak cheese | 1 | 0 | 0.0 | −0.82 (NA) |
| Total | 113 | 49 | 43.0 | −0.50 (0.42) |

*SD: standard deviation; MPN: most probable number; NA: not applicable.

30; 95% CI (50–85)) were starch metabolizers (possessing *amy* gene), and 60% (18/30; 95% CI (40–77)) were bile salt metabolizers (*bsh* gene). While screening the LAB for AMR genes such as *vanA*, *vanB*, *blaTEM*, *blaCTX-M*, *qnrA*, *qnrB*, *tetA*, and *tetB*, none of the LAB were found to carry these antibiotic resistance genes (Supplementary Materials, Table S2).

*3.6. Antibiotic Susceptibility Testing.* The antibiotic susceptibility testing by using the BD Phoenix-TM 100 automated ID/AST system indicated that all the LAB were resistant to teicoplanin and vancomycin.

Out of 53% ($n = 60$) isolated presumptive pathogens, 42% ($n = 46$) of the isolates were antibiotic resistance (including duplicates). Staphylococci 39% ($n = 42$) made up the majority of the resistant isolates and exhibited beta-lactamase (BLACT), methicillin-resistant *Staphylococcus* (MRS), high-level mupirocin (HLMUP), vancomycin-resistant*Staphylococcus aureus*, (VRSA) or *Staphylococcus* MLSb: macrolides, lincosamides, and streptogramin B (STAMLS) phenotype by antibiotic susceptibility testing.

After staphylococci, the other antibiotic-resistant isolates were *Enterococcus* spp. (2% ($n = 2$)), vancomycin-resistant enterococci (VRE), and *Providencia stuartii* (1% ($n = 1$)) with potential carbapenemase producer and *Micrococcus lylae* (1% ($n = 1$)) with nitrofurantoin resistance (Table 6) (Supplementary Materials, Table S1).

## 4. Discussion

In this study, we analyzed 113 fermented food samples and found 30 potential LAB isolates within four different genera and seven different species, identified as *Pediococcus pentosaceus*, *P. parvulus*, *P. acidilactici*, *P. damnosus*, *Leuconostoc mesenteroides*, *Aerococcus viridans*, and *Lactococcus plantarum*. Functional characterization of LAB isolates revealed that 80% of the LAB isolates were producers of riboflavin synthesis, 70% showed starch metabolism, and 60% possessed bile salt indicating their beneficial role in fermented food.

Amylolytic activity was observed in 33% of LAB by the phenotypic test, whereas the presence of the *amy* gene was observed in 70% of LAB by the molecular method, indicating higher sensitivity of the molecular method than the phenotypic test in detecting starch metabolism. Detection of the *amy* gene among LAB isolates indicates that these isolates were capable of sustaining growth in starchy substrates through the production of amylase [40].

None of the LAB were found to exhibit proteolytic activity; this could be due to absence or poor synthesis of proteinase enzyme in the isolated LAB. The proteolytic activity does not operate for many LAB [41], and this could be the reason that all the LAB in our study were negative for proteolytic activity.

In addition to probiotic effects, the LAB isolates revealed the potential for nutritional enhancement of foods by the production of riboflavin. The probiotic potential of LAB was

Table 5: Microbiological identification of LAB and pathogens (in duplicates) from fermented food samples.

| Food items | Number of fermented food samples containing LAB* | Isolated LAB in fermented food | Number of fermented food samples containing pathogens* | Isolated potential pathogens in fermented food |
|---|---|---|---|---|
| Fermented dry fish ($n = 29$) | 11 | *Pediococcus acidilactici* (1), *Pediococcus parvulus* (10) | 44 | *Bacillus cereus* (3), *B. circulans* (1), *B. megaterium* (1), *B. pumilus* (1), *B. subtilis* (2), *Dermacoccus nishinomiyaensis* (1), *Enterococcus faecium* (1), *Micrococcus lylae* (1), *Staphylococcus capitis* (3), *S. auricularis* (2), *S. cohnii* (6), *S. epidermidis* (1), *S. equorum* (1), *S. hominis* (4), *S. kloosii* (1), *S. pasteuri* (3), *S. pettenkoferi* (1), *S. saprophyticus* (2), *S. vitulinus* (1), *S. warneri* (4), *S. xylosus* (3), *Streptococcus acidominimus* (1) |
| Fermented bamboo shoots ($n = 43$) | 18 | *Aerococcus viridans* (1), *Leuconostoc mesenteroides* (3), *Pediococcus acidilactici* (4), *Pediococcus damnosus* (1), *Pediococcus pentosaceus* (9) | 38 | *Bacillus megaterium* (1), *B. subtilis* (1), *Burkholderia gladioli* (1), *Enterococcus casseliflavus/gallinarum* (1), *E. faecalis* (1), *E. faecium* (3), *Lysinibacillus sphaericus* (4), *Pasteurella aerogenes* (2), *Providencia stuartii* (1), *Serratia plymuthica* (1), *Staphylococcus warneri* (1), *S. aureus* (2), *S. cohnii spp. urealyticum* (1), *S. epidermidis* (3), *S. haemolyticus* (1), *S. lentus* (2), *S. sciuri* (1), *S. warneri* (1), *Streptococcus acidominimus* (5), *S. bovis* II (2), *S. porcinus* (3) |
| Fermented soyabeans ($n = 36$) | 1 | *Lactococcus plantarum* (1) | 59 | *Alloiococcus otidis* (1), *Bacillus cereus* (2), *B. circulans* (1), *B. pumilus* (1), *Corynebacterium matruchotii* (2), *Enterococcus faecalis* (2), *E. faecium* (16), *E. raffinosus* (1), *Proteus mirabilis* (1), *P. vulgaris* (3), *Providencia rettgeri* (1), *Pseudomonas putida* (1), *Serratia marcescens* (1), *Staphylococcus aureus* (2), *S. epidermidis* (2), *S. haemolyticus* (1), *S. intermedius* (2), *S. lentus* (2), *S. pettenkoferi* (1), *S. saprophyticus* (3), *S. sciuri* (1), *Streptococcus acidominimus* (1), *S. agalactiae* (2), *S. bovis* II (6), *S. porcinus* (3) |
| Fish pickles ($n = 1$) | 0 | — | 0 | 0 |
| Meat pickles ($n = 1$) | 0 | — | 0 | 0 |
| Milk products ($n = 2$) | 0 | — | 0 | 0 |
| Yak cheese ($n = 1$) | 0 | — | 4 | *Streptococcus bovis* II (1), *Staphylococcus capitis* (1), *S. haemolyticus* (1), *S. intermedius* (1) |
| Total | 30 | — | 145 | — |

*Including duplicates.

Table 6: Pathogens with antibiotic-resistant markers in fermented food.

| Fermented food | Pathogens | Resistance markers* |
|---|---|---|
| Bamboo shoot | *Staphylococcus epidermidis* | BLACT, MRS |
| Bamboo shoot | *Staphylococcus aureus* | BLACT |
| Bamboo shoot | *Staphylococcus haemolyticus* | MRS, HLMUP |
| Bamboo shoot | *Staphylococcus lentus* | BLACT, MRS |
| Bamboo shoot | *Staphylococcus sciuri* | MRS |
| Bamboo shoot | *Enterococcus faecalis* | VRE |
| Bamboo shoot | *Providencia stuartii* | Potential carbapenemase producer |
| Bamboo shoot | *Staphylococcus cohnii* ssp. *Urealyticum* | BLACT, MRS |
| Bamboo shoot | *Staphylococcus epidermidis* | BLACT, MRS |
| Bamboo shoot | *Staphylococcus aureus* | BLACT |
| Dry fish | *Staphylococcus cohnii* ssp. *Cohnii* | BLACT |
| Dry fish | *Staphylococcus hominis* | BLACT, MRS, HLMUP |
| Dry fish | *Staphylococcus pasteuri* | BLACT, MRS |
| Dry fish | *Staphylococcus xylosus* | BLACT, MRS |
| Dry fish | *Staphylococcus equorum* | BLACT |
| Dry fish | *Staphylococcus vitulinus* | BLACT, MRS |
| Dry fish | *Staphylococcus pettenkoferi* | BLACT, MRS |
| Dry fish | *Micrococcus lylae* | Nitrofurantoin resistance |
| Dry fish | *Staphylococcus xylosus* | BLACT |
| Dry fish | *Staphylococcus cohnii* ssp *urealyticum* | BLACT, MRS |
| Dry fish | *Staphylococcus xylosus* | BLACT, MRS |
| Dry fish | *Staphylococcus capitis* | BLACT, MRS |
| Dry fish | *Staphylococcus cohnii* ssp. *Cohnii* | BLACT |
| Dry fish | *Staphylococcus hominis* | BLACT, MRS, HLMUP |
| Dry fish | *Staphylococcus saprophyticus* | BLACT, STAMLS |
| Dry fish | *Staphylococcus cohnii* ssp. *Cohnii* | BLACT |
| Dry fish | *Staphylococcus hominis* | BLACT, MRS |
| Dry fish | *Staphylococcus pasteuri* | STAMLS *Staphylococcus* MLSb phenotype |
| Dry fish | *Staphylococcus saprophyticus* | BLACT, STAMLS |
| Dry fish | *Staphylococcus cohnii* ssp. *Cohnii* | BLACT |
| Dry fish | *Staphylococcus hominis* | BLACT, MRS |
| Dry fish | *Staphylococcus pasteuri* | STAMLS *Staphylococcus* MLSb phenotype |
| Soyabean | *Staphylococcus saprophyticus* | BLACT, MRS |
| Soyabean | *Staphylococcus lentus* | BLACT, MRS |
| Soyabean | *Staphylococcus aureus* | BLACT, MRS, HLMUP, STAMLS |
| Soyabean | *Staphylococcus intermedius* | BLACT, MRS, HLMUP, STAMLS |
| Soyabean | *Staphylococcus epidermidis* | BLACT, MRS, HLMUP, STAMLS |
| Soyabean | *Staphylococcus intermedius* | BLACT, MRS, HLMUP, STAMLS |
| Soyabean | *Staphylococcus aureus* | BLACT, MRS |
| Soyabean | *Staphylococcus sciuri* | MRS |
| Soyabean | *Staphylococcus epidermidis* | BLACT, MRS, HLMUP, STAMLS |
| Soyabean | *Enterococcus faecium* | VRE |
| Soyabean | *Staphylococcus saprophyticus* | BLACT, MRS |
| Soyabean | *Staphylococcus lentus* | BLACT, MRS |
| Yak cheese | *Staphylococcus haemolyticus* | BLACT, MRS, HLMUP, STAMLS |
| Yak cheese | *Staphylococcus intermedius* | BLACT, MRS, HLMUP, STAMLS |

*BLACT: beta-lactamase producer; MRS: methicillin-resistant *Staphylococcus*; HLMUP: high-level mupirocin resistance; STAMLS: macrolide-lincosamide-streptogramin-resistant *Staphylococcus*; STAMLS: *Staphylococcus* MLSb: macrolides, lincosamides, and streptogramin B.

further strengthened by their tolerance to bile salt, which might ensure their survival inside the gastrointestinal tract. Our results indicated a great diversity of LAB among the traditional fermented foods of Northeastern India with probiotic potential, but interestingly, in most of the food samples, LAB were absent and in fact had potential pathogens that sometimes carried antibiotic resistance.

Although there are several earlier studies about bacterial diversity in fermented foods [42, 43], we still know very little about the antimicrobial resistance implications on the safety of these foods in India. Our study found that all the LAB were resistant to antibiotics such as teicoplanin and vancomycin. The resistance to glycopeptides such as teicoplanin and vancomycin should be a point of concern as vancomycin is a last resort antibiotic in treating infections associated with multidrug-resistant Gram-positive bacteria [44, 45]. Though LAB have been reported to be intrinsically resistant to a number of antibiotics [46, 47], however in our study, none of the LAB were positive for AMR genes (*VanA, VanB, TEM, CTX, Qnr, TetA, and TetB*) by the molecular method,

indicating the absence of transferable antibiotic resistance genes [48, 49] and their safety with respect to AMR.

Assessment of food safety parameters revealed a wide variation in APC among the samples. This variation may be attributable to hygienic standards followed in the preparation of foods, but other factors such as the physiochemical nature of the food could also influence the likelihood of the food product to be a good medium for bacterial growth [50].

Coliforms were present in 43% of the fermented food samples, indicating poor hygienic standards. The majority (53%) of the fermented food samples were contaminated with different food pathogens, among which staphylococci were most dominant. This indicates a possible risk of foodborne disease from fermented foods. However, some foods are cooked before consumption [51–53], and some studies have even suggested that fermented food be cooked prior to consumption [18, 54] to reduce the risk of foodborne outbreaks. Nonetheless, episodes of food poisoning due to the consumption of traditional fermented food in India, such as Hawaijar, are occasionally reported [55], and there have been reports of food poisoning from fermented food products in other parts of the world as well [20, 23].

Staphylococci were discovered to be the most prevalent pathogens, and the prevalence of antibiotic-resistant bacterial pathogens was found to be 42%. Some of the staphylococci showed resistance against many classes of antimicrobials including beta-lactam, gentamicin, macrolides, and others. The major phenotypic resistance markers observed in staphylococci were BLACT, MRS, HLGR, HLMuRS, mecA-RS, and Staph MLSb. After staphylococci, other antibiotic-resistant pathogens identified were *Enterococcus* spp., *Providencia stuartii*, and *Micrococcus lylae* that were vancomycin-resistant enterococci (VRE), potential carbapenemase producers, and nitrofurantoin resistant.

The presence of antimicrobial resistance traits among isolates in fermented food indicates potential risks to consumers due to the transfer of antimicrobial resistance [56] to other microorganisms [57] in the gastrointestinal tract and may lead to possible treatment failure of infections [58].

According to the World Health Organization (WHO), food-handling personnel plays an important role in ensuring food safety throughout the chain of food production, processing, storage, and preparation [59]. Mishandling and disregarding hygienic measures on the part of food vendors may enable pathogens to contaminate the food [60] and, in some cases, to survive and multiply in sufficient numbers to cause illness to consumers [61]. Traditional fermented foods occupy an important food niche in Northeast India [16]. The fermentation process is an alternative method of food preservation of raw food and thus helps in increasing the shelf life [62] of the food product. Fermented food influences the gut microbiota of consumers and could have potential health advantages [15, 63].

Our study was limited by the small number of samples, particularly from some regions, and the wide variety of products. The differences in the quantity of the collected fermented food samples were caused due to seasonal variations in the availability of raw material and reduced local production of fermented food. Given that the study is not representative, the prevalence of food pathogens in the samples was less conclusive than would have been the case with a more representative sampling frame. The virulence of the pathogenic isolates in fermented food is also unknown because a molecular approach was not used to confirm the pathogenicity of the isolates. Therefore, extensive and systemic future studies are required with larger sample frames in order to understand the actual scenario with regard to the safety of traditional fermented food products in Northeast India.

## 5. Conclusions

Our findings revealed a mixed picture of opportunity, with a large variety of LAB with probiotic potential on the one hand, and a significant risk, with poor hygienic quality, the presence of pathogens, and the prevalence of drug resistance traits on the other. This is the first study of LAB in conjunction with foodborne bacterial pathogens and their antibiotic resistance in traditional fermented foods found in Northeast India. This was, however, only a small study, and more extensive studies are required with larger samples in order to understand the actual scenario with regard to the safety of traditional fermented food products and the role of LAB.

## Data Availability

Data are made available from the authors upon reasonable request.

## Conflicts of Interest

The authors declare no conflicts of interest.

## Authors' Contributions

S.G and T.K.D were responsible for conceptualization. S.G and T.K.D were responsible for methodology. T.K.D and S.G were responsible for validation. T.K.D was responsible for formal analysis. T.K.D was responsible for investigation. S.G and T.K.D were responsible for resources. T.K.D, S.G, and J.F.L were responsible for writing the original draft. T.K.D, J.F.L, S.G, Å.L, R.S, A.A.P.M, S.D, P.K, and A.S were responsible for writing, reviewing, and editing the manuscript. J.F.L, T.K.D, S.G, and Å.L were responsible for visualization. S.G and J.F.L were responsible for supervision. S.G was responsible for project administration. T.K.D and S.G were responsible for funding acquisition. All the authors have read and agreed to the published version of the manuscript.

## Acknowledgments

The authors thankfully acknowledge funding and laboratory support received from the Division of Animal and Fisheries Sciences, Indian Council for Agricultural Research, Northeastern Hill Region, Meghalaya, India. The authors also thankfully acknowledge the support received from the scientists and colleagues from the Division of Animal and

Fisheries Sciences, Indian Council for Agricultural Research, Meghalaya, India, for helping in sample collection.

## Supplementary Materials

The following supporting information can be downloaded at https://www.hindawi.com. Table S1: details of fermented foods, including their aerobic plate count (APC), most probable number (MPN), associated pathogens, and resistance markers for those pathogens and Table S2: details of fermented foods and lactic acid bacteria (LAB) identification with functional and antibiotic resistance genes. (*Supplementary Materials*)

## References

[1] N. Şanlier, B. B. Gökcen, and A. C. Sezgin, "Health benefits of fermented foods," *Critical Reviews in Food Science and Nutrition*, vol. 59, no. 3, pp. 506–527, 2019.

[2] R. Sharma, P. Garg, P. Kumar, S. K. Bhatia, and S. Kulshrestha, "Microbial fermentation and its role in quality improvement of fermented foods," *Fermentation*, vol. 6, no. 4, p. 106, 2020.

[3] O. Adolfsson, S. N. Meydani, and R. M. Russell, "Yogurt and gut function," *The American Journal of Clinical Nutrition*, vol. 80, no. 2, pp. 245–256, 2004.

[4] N. K. Leeuwendaal, C. Stanton, P. W. O'Toole, and T. P. Beresford, "Fermented foods, health and the gut microbiome," *Nutrients*, vol. 14, no. 7, p. 1527, 2022.

[5] A. Zapaśnik, B. Sokołowska, and M. Bryła, "Role of lactic acid bacteria in food preservation and safety," *Foods*, vol. 11, no. 9, p. 1283, 2022.

[6] M. E. Margulis, "The regime complex for food security: implications for the global hunger challenge," *Global Governance*, vol. 19, p. 53, 2013.

[7] G. Wu, J. Fanzo, D. D. Miller et al., "Production and supply of high-quality food protein for human consumption: sustainability, challenges, and innovations," *Annals of the New York Academy of Sciences*, vol. 1321, no. 1, pp. 1–19, 2014.

[8] O. K. Achi, "The potential for upgrading traditional fermented foods through biotechnology," *African Journal of Biotechnology*, vol. 4, no. 5, pp. 375–380, 2005.

[9] K. C. Anukam and G. Reid, "African traditional fermented foods and probiotics," *Journal of Medicinal Food*, vol. 12, no. 6, pp. 1177–1184, 2009.

[10] W. H. Holzapfel, "Appropriate starter culture technologies for small-scale fermentation in developing countries," *International Journal of Food Microbiology*, vol. 75, no. 3, pp. 197–212, 2002.

[11] E. M. Selhub, A. C. Logan, and A. C. Bested, "Fermented foods, microbiota, and mental health: ancient practice meets nutritional psychiatry," *Journal of Physiological Anthropology*, vol. 33, no. 1, p. 2, 2014.

[12] W. Sybesma, R. Kort, and Y. K. Lee, "Locally sourced probiotics, the next opportunity for developing countries?" *Trends in Biotechnology*, vol. 33, no. 4, pp. 197–200, 2015.

[13] V. Bell, J. Ferrão, and T. Fernandes, "Nutritional guidelines and fermented food frameworks," *Foods*, vol. 6, no. 8, pp. 1–17, 2017.

[14] S. Sekar and S. Mariappan, "Usage of traditional fermented products by Indian rural folks and IPR," *Indian Journal of Traditional Knowledge*, vol. 6, no. 1, pp. 111–120, 2007.

[15] M. L. Marco, D. Heeney, S. Binda et al., "Health benefits of fermented foods: microbiota and beyond," *Current Opinion in Biotechnology*, vol. 44, pp. 94–102, 2017.

[16] M. R. Swain, M. Anandharaj, R. C. Ray, and R. Parveen Rani, "Fermented fruits and vegetables of asia: a potential source of probiotics," *Biotechnology Research International*, vol. 2014, Article ID 250424, 19 pages, 2014.

[17] C. R. Kok and R. Hutkins, "Yogurt and other fermented foods as sources of health-promoting bacteria," *Nutrition Reviews*, vol. 76, pp. 4–15, 2018.

[18] M. J. R. Nout, "Fermented foods and food safety," *Food Research International*, vol. 27, no. 3, pp. 291–298, 1994.

[19] Y. Motarjemi and M. J. R. Nout, "Update Le point Food fermentation: a safety and nutritional," *Bulletin of the World Health Organization*, vol. 74, no. 6, pp. 553–559, 1996.

[20] N. Benkerroum, "Traditional fermented foods of North African countries: technology and food safety challenges with regard to microbiological risks," *Comprehensive Reviews in Food Science and Food Safety*, vol. 12, no. 1, pp. 54–89, 2013.

[21] S. Keisam, N. Tuikhar, G. Ahmed, and K. Jeyaram, "Toxigenic and pathogenic potential of enteric bacterial pathogens prevalent in the traditional fermented foods marketed in the Northeast region of India," *International Journal of Food Microbiology*, vol. 296, pp. 21–30, 2019.

[22] L. Topisirovic, M. Kojic, D. Fira, N. Golic, I. Strahinic, and J. Lozo, "Potential of lactic acid bacteria isolated from specific natural niches in food production and preservation," *International Journal of Food Microbiology*, vol. 112, no. 3, pp. 230–235, 2006.

[23] G. Zhou, K. Bester, B. Liao, Z. Yang, R. Jiang, and N. B. Hendriksen, "Characterization of three Bacillus cereus strains involved in a major outbreak of food poisoning after consumption of fermented black beans (Douchi) in Yunan, China," *Foodborne Pathogens and Disease*, vol. 11, no. 10, pp. 769–774, 2014.

[24] B. Tamang, J. P. Tamang, U. Schillinger, C. M. A. P. Franz, M. Gores, and W. H. Holzapfel, "Phenotypic and genotypic identification of lactic acid bacteria isolated from ethnic fermented bamboo tender shoots of North East India," *International Journal of Food Microbiology*, vol. 121, no. 1, pp. 35–40, 2008.

[25] P. J. Quinn, B. K. Markey, and F. C. Leonard, "Concise review of veterinary microbiology," 2015, http://ebookcentral.proquest.com/lib/uu/detail.action?docID=2050943.

[26] J. T. P. Larry Maturin, "Bacteriological analysis manual of food and drug administration," *Food and Drug Administration (FDA)*, Silver Spring, Berlin, Germany, 2001, https://www.fda.gov/food/laboratory-methods-food/bam-chapter-3-aerobic-plate-count, 8th edition.

[27] P. Feng and S. D. Weagant, "Enumeration of *Escherichia coli* and the coliform bacteria," *Food and Drug Administration (FDA)*, Silver Spring, Berlin, Germany, 2002, https://www.fda.gov/food/laboratory-methods-food/bam-chapter-4-enumeration-escherichia-coli-and-coliform-bacteria, 8th edition.

[28] S. Tallent and J. Hait, "Bam: staphylococcus aureus. u.s. food and drug administration," 2019, https://www.fda.gov/food/laboratory-methods-food/bam-chapter-12-staphylococcus-aureus.

[29] J. Beganović, B. Ž. Kos, A. Leboš Pavunc, K. Uroić, P. Džidara, and J. Šušković, "Proteolytic activity of probiotic strain Lactobacillus helveticus M92," *Anaerobe*, vol. 20, pp. 58–64, 2013.

[30] S. Mayrhofer, A. H. A. M. Van Hoek, C. Mair et al., "Antibiotic susceptibility of members of the Lactobacillus acidophilus group using broth microdilution and molecular identification of their resistance determinants," *International Journal of Food Microbiology*, vol. 144, no. 1, pp. 81–87, 2010.

[31] V. Ocaña, C. Silva, and M. E. Nader-Macías, "Antibiotic susceptibility of potentially probiotic vaginal lactobacilli," *Infectious Diseases in Obstetrics and Gynecology*, vol. 2006, 2006.

[32] R. Temmerman, "Identification and antibiotic susceptibility of bacterial isolates from probiotic products," *International Journal of Food Microbiology*, vol. 81, p. 1, 2003.

[33] Clsi, "CLSI document M100-S22; performance standards for antimicrobial susceptibility testing; twenty-second informational supplement," in *Clinical and Laboratory Standards Institute*, P. A. Wayne, Ed., vol. 32, no. 3, 2012.

[34] W. Turpin, C. Humblot, and J.-P. Guyot, "Genetic screening of functional properties of lactic acid bacteria in a fermented pearl millet slurry and in the metagenome of fermented starchy foods," *Applied and Environmental Microbiology*, vol. 77, no. 24, pp. 8722–8734, 2011.

[35] Y. Sáenz, L. Briñas, E. Domínguez et al., "Mechanisms of resistance in multiple-antibiotic-resistant*Escherichia coli* strains of human, animal, and food origins," *Antimicrobial Agents and Chemotherapy*, vol. 48, no. 10, pp. 3996–4001, 2004.

[36] L. M. Cavaco, N. Frimodt-Møller, H. Hasman, L. Guardabassi, L. Nielsen, and F. M. Aarestrup, "Prevalence of quinolone resistance mechanisms and associations to minimum inhibitory concentrations in quinolone-resistant*Escherichia coli* isolated from humans and swine in Denmark," *Microbial Drug Resistance*, vol. 14, no. 2, pp. 163–169, 2008.

[37] V. Cattoir, F. X. Weill, L. Poirel, L. Fabre, C. J. Soussy, and P. Nordmann, "Prevalence of qnr genes in Salmonella in France," *Journal of Antimicrobial Chemotherapy*, vol. 59, no. 4, pp. 751–754, 2007.

[38] S. Gröbner, D. Linke, W. Schütz et al., "Emergence of carbapenem-non-susceptibleextended-spectrum beta-lactamase-producing*Klebsiella pneumoniae* isolates at the university hospital of Tübingen, Germany," *Journal of Medical Microbiology*, vol. 58, pp. 912–922, 2009.

[39] Food Standards Australia New Zealand, "Compendium of microbiological criteria for food," in *Compendium Of Microbiological Criteria For Food (Issue October)*Food Standards Australia New Zealand Publishing, Canberra, Australia, 2016.

[40] E. H. Tou, J. P. Guyot, C. Mouquet-Rivier et al., "Study through surveys and fermentation kinetics of the traditional processing of pearl millet (Pennisetum glaucum) into bensaalga, a fermented gruel from Burkina Faso," *International Journal of Food Microbiology*, vol. 106, no. 1, pp. 52–60, 2006.

[41] M. Kojic, D. Fira, A. Banina, and L. Topisirovic, "Characterization of the cell wall-bound proteinase of lactobacillus casei HN14," *Applied and Environmental Microbiology*, vol. 57, pp. 1753–1757, 1991.

[42] S. De Mandal, S. S. Singh, R. B. Muthukumaran, K. Thanzami, V. Kumar, and N. S. Kumar, "Metagenomic analysis and the functional profiles of traditional fermented pork fat 'sa-um' of Northeast India," *AMB Express*, vol. 8, no. 1, 2018.

[43] N. Thapa, J. Pal, and J. P. Tamang, "Microbial diversity in ngari, hentak and tungtap, fermented fish products of North-East India," *World Journal of Microbiology and Biotechnology*, vol. 20, no. 6, pp. 599–607, 2004.

[44] M. S. Butler, K. A. Hansford, M. A. T. Blaskovich, R. Halai, and M. A. Cooper, "Glycopeptide antibiotics: back to the future," *Journal of Antibiotics*, vol. 67, no. 9, pp. 631–644, 2014.

[45] G. L. French, "The continuing crisis in antibiotic resistance," *International Journal of Antimicrobial Agents*, vol. 36, 2010.

[46] D. Billot-Klein, L. Gutmann, S. Sable, E. Guittet, and J. Van Heijenoort, "Modification of peptidoglycan precursors is a common feature of the low-level vancomycin-resistant VANB-type Enterococcus D366 and of the naturally glycopeptide-resistant species Lactobacillus casei, Pediococcus pentosaceus, Leuconostoc mesenteroides, and," *Journal of Bacteriology*, vol. 176, no. 8, pp. 2398–2405, 1994.

[47] S. Handwerger, M. J. Pucci, K. J. Volk, J. Liu, and M. S. Lee, "Vancomycin-resistant Leuconostoc mesenteroides and Lactobacillus casei synthesize cytoplasmic peptidoglycan precursors that terminate in lactate," *Journal of Bacteriology*, vol. 176, no. 1, pp. 260–264, 1994.

[48] B. G. Elisha and P. Courvalin, "Analysis of genes encoding D-alanine: D-alanine ligase-related enzymes in Leuconostoc mesenteroides and Lactobacillus spp," *Gene*, vol. 152, no. 1, pp. 79–83, 1995.

[49] F. Huygens, J. Knauer, T. Hecht, and P. Britz, "Vancomycin resistance is not inducible in Leuconostoc, Lactobacillus, Pediococcus and *Enterococcus faecalis* A 256," *South African Journal of Science*, vol. 91, no. 2, pp. 94–97, 1995.

[50] T. A. Hussein and J. K. Isa, "Study Of The Microbiological Quality And Physio-Chemical Properties Of The Soft White Cheese Traditionally Produced In Wasit Province In Iraq," *Nveo-Natural Volatiles & Essential Oils Journal Nveo*, vol. 8, pp. 48–61, 2021.

[51] S. Kose and G. M. Hall, "Sustainability of fermented fish products," *Fish Processing: Sustainability and New Opportunities*, pp. 138–166, 2010.

[52] R. K. Majumdar and S. Basu, "Characterization of the traditional fermented fish product Lona ilish of Northeast India," *Indian Journal of Traditional Knowledge*, vol. 9, no. 3, pp. 453–458, 2010.

[53] V. Waisundara, N. Jayawardena, and M. Watawana, "Safety of fermented fish products," in *Regulating Safety of Traditional and Ethnic Foods*, pp. 149–168, Elsevier, Amsterdam, Netherlands, 2016.

[54] G. Campbell-Platt, "Fermented meats—a world perspective," in *Fermented Meats*, pp. 39–52, Springer, Berlin, Germany, 1995.

[55] K. Jeyaram, W. Romi, T. A. Singh, A. R. Devi, and S. S. Devi, "Bacterial species associated with traditional starter cultures used for fermented bamboo shoot production in Manipur state of India," *International Journal of Food Microbiology*, vol. 143, no. 1–2, pp. 1–8, 2010.

[56] M. E. Sanders, L. M. A. Akkermans, D. Haller et al., "Safety assessment of probiotics for human use," *Gut Microbes*, vol. 1, no. 3, pp. 164–185, 2010.

[57] E. Y. Furuya and F. D. Lowy, "Antimicrobial-resistant bacteria in the community setting," *Nature Reviews Microbiology*, vol. 4, no. 1, pp. 36–45, 2006.

[58] R. Capita and C. Alonso-Calleja, "Antibiotic-resistant bacteria: a challenge for the food industry," *Critical Reviews in Food Science and Nutrition*, vol. 53, no. 1, pp. 11–48, 2013.

[59] Who, "Health surveillance and management procedures for food-handling personnel: report of a WHO consultation," 1989, http://WHO_TRS_785.pdf.
[60] S. Rane, "Street vended food in developing world: hazard analyses," *Indian Journal of Microbiology*, vol. 51, no. 1, pp. 100–106, 2011.
[61] G. Girma, T. Ketema, and K. Bacha, "Microbial load and safety of paper currencies from some food vendors in jimma town, southwest ethiopia," *BMC Research Notes*, vol. 7, 2014.
[62] Y. Motarjemi, "Impact of small scale fermentation technology on food safety in developing countries," *International Journal of Food Microbiology*, vol. 75, no. 3, pp. 213–229, 2002.
[63] F. Melini, V. Melini, F. Luziatelli, A. G. Ficca, and M. Ruzzi, "Health-promoting components in fermented foods: an up-to-date systematic review," *Nutrients*, vol. 11, no. 5, p. 1189, 2019.