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Identification and antifungal potential of lactic acid bacteria strains isolated from *Attiéké* (*Cassava semolina*) ferment for tomato bio-preservation

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Abstract

Population growth in recent years has led to an increase in the consumption of fruits and vegetables. However, these plant-based foods are often prone to fungal contamination, which poses significant preservation challenges. This study aimed to identify lactic acid bacteria (LAB) strains from cassava ferment and evaluate their antifungal activity. In this study, LAB were isolated from cassava ferment and tested for their antifungal properties as well as their sensitivity to antibiotics. The LAB isolates exhibited antifungal activity with inhibition zones ranging from 1 mm to over 29 mm against various phytopathogenic moulds. They were selected and identified by 16s rRNA sequencing method. Nine strains demonstrated strong inhibitory activity against *Fusarium sp., Colletotrichum gloeosporioides, Botryodiplodia theobromae, Aspergillus carbonarius,* and *Aspergillus niger*. These selected LAB were sensitive to penicillin, rifampicin, imipenem, linezolid, clindamycin, and cefepime. Two species *Limosilactobacillus fermentum* and *Weissella* confuse were identified. Both supernatant and cells of these species exhibited good preservation of tomatoes at room temperature, the supernatant being the most effective. This study clearly demonstrates the potential of LAB strains from cassava fermented foods and highlights their potential applications in food bio-preservation.

Keywords: Lactic acid bacteria; *Limosilactobacillus fermentum*; *Weissella confusa*; Bio-preservation; Antifungal activity; Attieke ferment

1. Introduction

The consumption of fresh fruits and vegetables is important for human health. These foods help in reducing the risk of cardiovascular diseases and can also lower the incidence of certain cancers [1]. According to the WHO, inadequate intake of fruits and vegetables is responsible for around 19% of deaths from gastrointestinal cancers, 31% of deaths from ischemic heart disease, and 11% of fatal cardiovascular events worldwide [2]. However, the fresh fruit and vegetable industry faces several challenges, with shelf life being a major concern. Climacteric fruits are particularly perishable because, after harvesting, they continue to exhibit metabolic activity [3]. The primary factors limiting their shelf life are senescence, transpiration, and infections caused by bacteria such as *Pseudomonas* sp. and *Erwinia* sp. [4], as well as fungal infections caused by moulds like *Botrytis cinerea*, *Fusarium* sp., *Rhizopus* sp., *Alternaria alternata*, *Sclerotinia sclerotiorum*, *Colletotrichum gloeosporioides*, and *Penicillium* sp. These infections are widely implicated in post-harvest diseases and rots of fruits such as tomatoes, chilies, eggplants, papayas, and avocados [4]. More than 10% of the world's food production is lost due to mould proliferation in these crops, and the toxins produced by these fungi

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are considered as the most dangerous food [5]. Fungi are among the most significant pathogens affecting fruits and vegetables, causing serious diseases such as rot and gall. These spoilage microorganisms can be introduced during cultivation, crop growth in the field, harvesting, post-harvest handling, and storage and distribution [6]. Under the influence of environmental factors, they pose a significant threat to the production, marketing, and consumption of fresh fruit [7].

The deterioration of fruits and vegetables due to infections can be reduced by actively maintaining their natural resistance, delaying senescence, and minimizing mechanical injury [3]. However, these measures are often insufficient to provide adequate protection against infection. Therefore, the use of chemical fungicides is often necessary to control disease, ensuring a profitable market price for producers, and maintaining quality for consumers [8]. However, due to the potential carcinogenic, teratogenic, and allergenic effects of these chemicals, as well as their high residual toxicity for consumers, the use of chemical preservatives must be reduced. A promising alternative method is bio-preservation, which is gaining attention due to its efficacy [9] and its antagonistic properties, ubiquity, diversity, and persistence in the environment. Bio-preservation is a biological method that has been the subject of numerous studies over the last 40 years [10]. It enables the natural preservation of foods by maintaining their organoleptic and nutritional properties, which are often lost through chemical agents or heat treatments [11]. Bio-preservation involves extending the shelf life and improving the safety of food products such as fruits and vegetables using microorganisms and/or their metabolites [10]. Microorganisms used for bio-preservation must be safe for humans and animals. Among these, lactic acid bacteria (LAB) are well-known for their safety and antibacterial activity, which helps in reducing the use of chemicals in food [12]. The bacteriocins produced by LAB make them excellent preservatives for food products [13-14]. LAB and/or their metabolites have been used for millennia by many populations to contribute to food preservation [15].

This research study aimed to investigate the bio-preservation potential of LAB strains isolated from *attiéké* (*cassava semolina*) ferment, focusing on their antifungal and antibiotic activities, molecular identification, and the effectiveness of tomato bio-preservation through immersion and coating.

2. Material and methods

2.1. Lactic acid bacteria strains

From preliminary tests, nine lactic acid bacteria strains isolated from *attiéké* (cassava) ferment were selected. These strains were grown in MRS broth for 48 h at 37 °C.

2.2. Lactic acid bacteria isolation and identification

After cultivation in MRS broth for 48 h at 37 °C, lactic acid bacteria cells were harvested by centrifugation at 5000 x g for 10 min. Genomic DNA extraction was performed using a ZR Fungal/Bacterial DNA kit (Zymo Research, Irvine, CA, USA), according to the manufacturer's instructions. DNA concentration and purity were checked using a SpectraMax® QuickDrop™ (Molecular Devices, San Jose, CA, USA). LAB strains were identified by analysis of 16S rDNA amplified with universal primers 27F (AGAGTTTGATCMTGGCTCAG) and 1492R (TACGGTACCTTGTTACGACTT) (Biolegio B.V. Nijmegen, Netherlands). The reaction mixture contained 50 µL DreamTaq 10X green buffer (contains 20 mM MgCl2), 0.5 µM of each primer, 0.2 mM dNTP, 0.025 U DreamTaq DNA polymerase (Thermo Fisher Scientific, Baltics, UAB, Vilnius, Lithuania), and 10 ng bacterial DNA. The amplification protocol included an initial denaturation at 95°C for 3 minutes, followed by 35 cycles (94°C for 1 minute, 55 °C for 1 minute, 72 °C for 1 minute) and a final extension step at 72°C for 7 minutes. Amplification reactions of the 16S rDNA region were performed using a thermal cycler (MultiGene Thermal Cycler Labnet International, Inc., Edison, NJ, USA). PCR products were detected by agarose gel electrophoresis (2% (w/v) agarose, 90 V, 60 min) and visualized using a GelDoc-It imaging system (Analytik Jena, Upland, CA, USA). Sequencing was performed in both directions with universal primers 27F and 1492R by Cellular and Molecular Immunological Application, Holland (CEMIA, Greece). The partially obtained nucleotide sequences were aligned with multiple homologous sequences available in the GenBank databases of the National Center for Biotechnology Information (NCBI) (http://www.ncbi.nlm.nih.gov, accessed August 2023) to identify species based on high similarity. A phylogenetic tree was constructed using the neighbor-joining method [16] with Molecular Evolution Genetic Analysis (MEGA) software, version X [17].

2.3. Antifungal activity of selected lactic acid bacteria isolates

Antagonistic activities of lactic acid bacteria isolates (LAB) were evaluated against five spoilage-indicating moulds (*Fusarium* sp, *Colletotrichum gloesporioide*, *Botryodiplodia theobromae*, *Aspergillus carbonarius* and, *Aspergillus niger*) using the agar well diffusion method described by Balouiri et al. [18] with some modifications. These moulds were obtained from phytopathology and plant physiology Laboratory of University Nangui Abrogoua, Abidjan, Ivory Coast.

After 48 h of growth in MRS broth at 37 °C, the lactic acid bacteria (LAB) isolates were centrifuged for five minutes at 10,000 rpm and 4°C. Cell-free supernatants (CFS) were obtained by filtering the supernatant through sterile 0.22 µm Millipore filters. Fresh cultures of the indicator moulds (approximately 10^7 CFU/mL) were added to potato dextrose agar (PDA) medium, which was then solidified in petri dishes. Wells with a diameter of 6 mm were aseptically punched into the agar using a sterile tip, filled with 100 µL of CFS, and incubated for 24 hours at 37 °C. The diameter of the inhibition zones surrounding each well was measured in millimeters (mm). A clear inhibition zone of 1 mm or more was considered indicative of the inhibitory activity of the LAB isolates.

2.4. Antibiotics Susceptibility Test

Antibiotic susceptibility testing was performed using the disc diffusion method with antibiotic-impregnated discs. After adjusting the inoculum density to an Optical Density (OD) of 0.5 at 600nm (10^7 CFU/mL), the MRS medium was inoculated and allowed to dry. Thirteen different commercial antibiotic disks were applied to plate. The antibiotics used were Penicillin G (10 µg), Oxacillin (1 µg), Rifampicin (5 µg), Kanamycin (30 µg), Ciprofloxacin (5 µg), Imipenem (10 µg), Vancomycin (30 µg), Amikacin (30 µg), Linezolide (30 µg), Clindamycin (2 µg), Sulfamethoxazole (23.75 µg), Fusidic acid (10 µg), Cefepime (30 µg). The antibiotic discs used were of Becton Dickson marks (USA). After incubating at 30 °C for 24 h under anaerobic conditions, zones of inhibition were observed around the discs and the diameter of the inhibition zone was measured. Lactic acid bacteria (LAB) strains were classified according to the Clinical and Laboratory Standards Institute [19] guidelines as: Resistant (R): Diameter ≤ 14 mm, Intermediate Resistant (IR): Diameter between 15 and 19 mm, Sensitive (S): Diameter > 19 mm.

2.5. Bio-preservation Test on Tomatoes by Immersion and Coating

The two strains with the highest antifungal activity were used for the bio-preservation test. The selected lactic acid bacteria (LAB) strains were grown in MRS broth for 48 hours at 37 °C after inoculation at an OD of 0.5 at 600 nm (10^7 CFU/mL). The cells and supernatants were separated by refrigerated centrifugation at 4 °C at 5000 x g during 10 min. Previously, tomato samples were contaminated with *Penicillium* sp. The harvested cells were used for bio-preservation by coating the surface of the tomatoes, while the supernatants were used for bio-preservation by immersion, achieved by plunging the tomatoes for 10 minutes. Untreated tomatoes were used as negative controls. The cells and supernatants were applied individually and in combination. The tomatoes were stored at room temperature for 2 weeks, and their visual appearance was documented through photographs.

3. Results

3.1. Identification of selected lactic acid bacteria strains

The sequencing of the selected LAB isolates followed by BLAST analysis showed that all isolates belonged to two species *Limosilactobacillus fermentum* and *Weissella confusa*. The partial DNA sequences of all strains have been deposited in the NCBI database, with accession numbers OR458563–OR458571. The species names and corresponding details are provided in Table 1. In addition, the phylogenetic tree revealed kinship links between the study strains and those in the GenBank database (Figure 1*). Limosilactobacillus fermentum* strains B5, B9, B27, and B33 were closely related to *Limosilactobacillus fermentum* TMPC 46B23. Similarly, *Weissella confusa* strains B17 and B22 were linked to *Weissella confusa* TMPC 40915 and *Weissella confusa* 1010. Interestingly, *Limosilactobacillus fermentum* B13 in this study showed a relationship with *Weissella confusa* strains B10 and B35, also from this study.

Table 1 Identification of Lactic Acid Bacteria Strains and their Accession Numbers

Isolates	Species	Accession Numbers
B5	*Limosilactobacillus fermentum*	OR458563
B9	*Limosilactobacillus fermentum*	OR458564
B10	*Weissella confusa*	OR458565
B13	*Limosilactobacillus fermentum*	OR458566
B17	*Weissella confusa*	OR458567
B22	*Weissella confusa*	OR458568
B27	*Limosilactobacillus fermentum*	OR458569

B33	*Limosilactobacillus fermentum*	OR458570
B35	*Weissella confusa*	OR458571

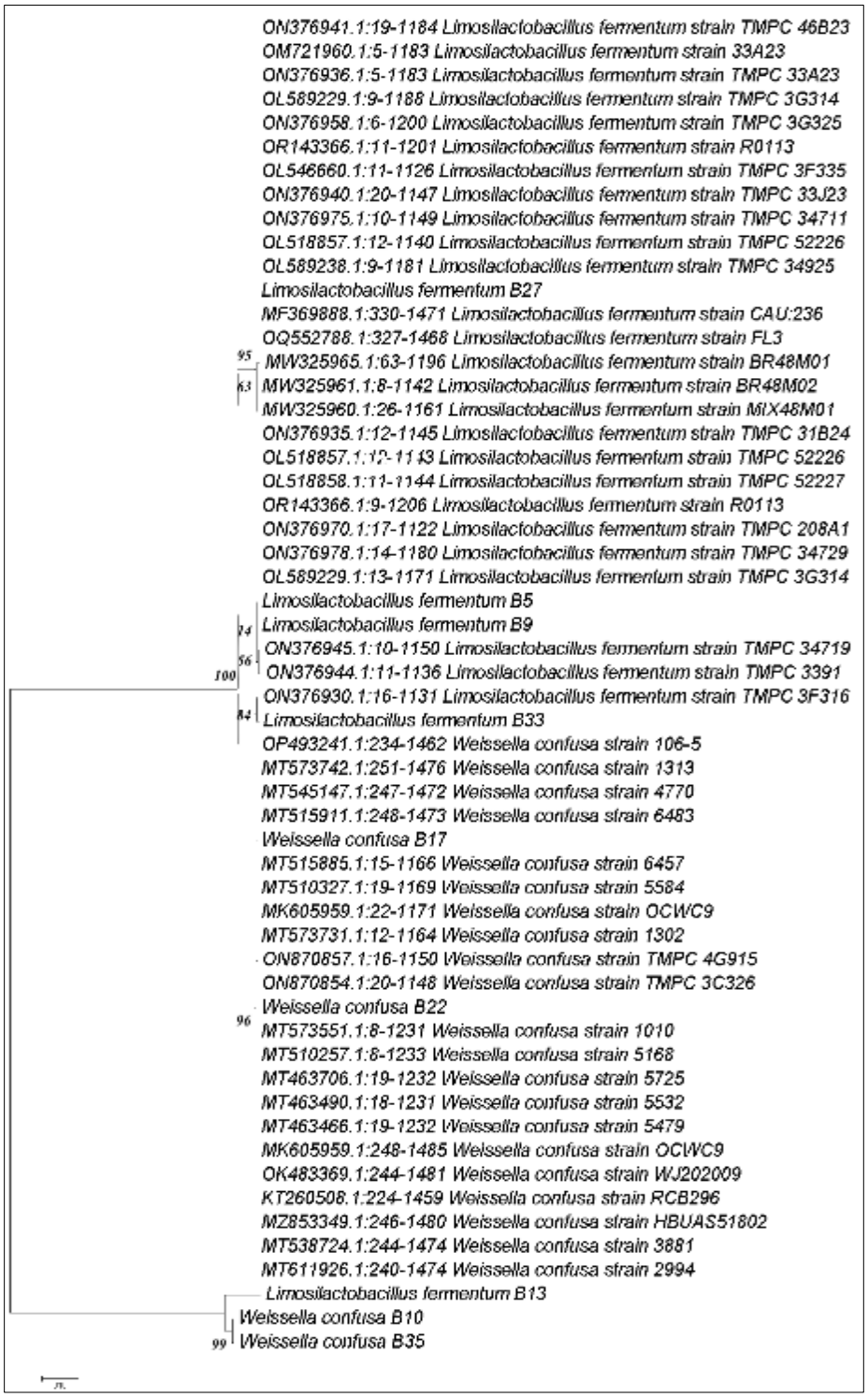

Figure 1 The phylogenetic tree showing the relative position of *Limosilactobacillus fermentum* and *Weissella confusa* species as inferred by the neighbor-joining method with 16S rRNA gene sequences

3.2. Antifungal activity of lactic acid bacteria strains

Antifungal activity against phytopathogenic moulds is considered as the main criterion for selecting lactic acid bacteria strains with bio-preservation potential. The antifungal activity of lactic acid bacteria strains was assessed using the agar well diffusion method against five phytopathogenic fungal strains. The inhibitory effect of these strains was indicated by the appearance of a clear halo around the well, while the absence of a halo signified a lack of inhibitory activity (Figure 2). The diameters of the inhibition zones are presented in Table 2. Among the tested strains, strain B10 exhibited high antifungal activity against all phytopathogenic moulds. In contrast, strains B13 and B35 demonstrated high antifungal activity against most moulds, except *Fusarium* sp. and *Aspergillus niger*, respectively.

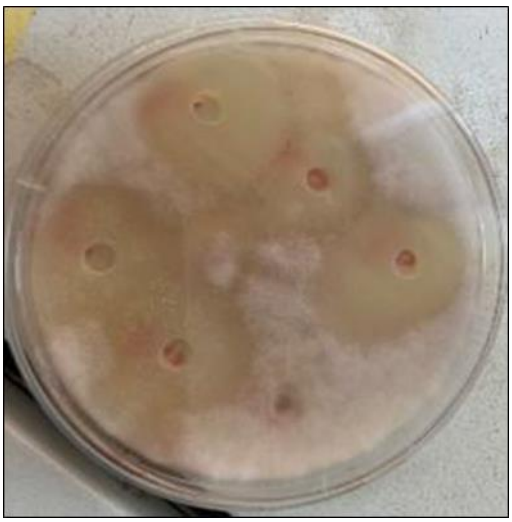

Figure 2 Antifungal activity of a lactic acid bacteria strain

Table 2 Diameters of the inhibition halos of lactic acid bacteria isolates

Strains	*Aspergillus carbonarius*	*Aspergillus niger*	*Fusarium sp*	*Colletotrichum gloesporioide*	*Botryodiplodia theobromae*
B5	+++	++	++	++	+++
B9	+++	++	+++	++	+++
B10	+++	+++	+++	+++	+++
B13	+++	++++	++	++++	++++
B17	+++	+++	+++	+++	+++
B22	+	+++	++++	++	+++
B27	++	+++	++	++	++
B33	+++	+++	++	+	+++
B35	++++	++	++++	++++	+++

(+) : 1–5 mm ; (+ +) : 6–17 mm ; (+ ++) : 18–29 mm (+++) ; 29 and more (++++)

3.3. Antibiogram of lactic acid bacteria isolates

Antibiotic susceptibility testing was carried out using the disk diffusion method (Figure 3). For this test, 13 antibiotics were used: Penicillin G (6 µg), Oxacillin (5 µg), Rifampicin (30 µg), Kanamycin (1 mg), Ciprofloxacin (5 µg), Imipenem (10 µg), Vancomycin (10 µg), Amikacin (10 µg), Linezolid (30 µg), Clindamycin (2 µg), Sulfamethoxazole (100 µg), Fusidic acid (10 µg), and Cefepime (30 µg). Generally, all strains were sensitive to Penicillin G, Rifampicin, Imipenem, Linezolid, Clindamycin, and Cefepime, and also exhibited multidrug resistance to Kanamycin and Sulfamethoxazole (Table 2).

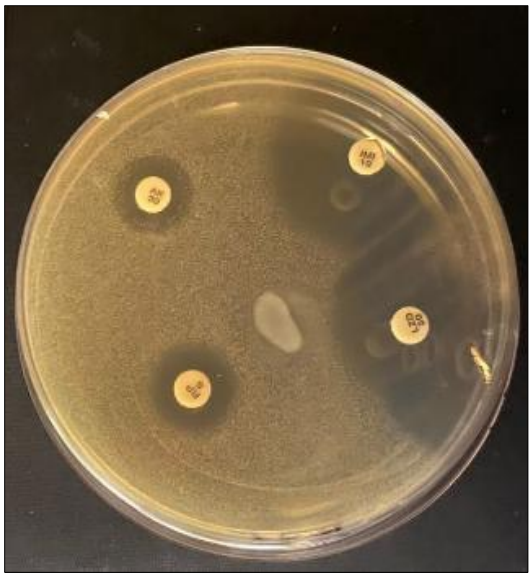

Figure 3 Antibiogram of a lactic acid bacteria strain

Table 3 Antibiotic susceptibility pattern of lactic acid bacteria strains

Strains	IMI	AK	CIP	LZD	K	RP	CD	OX	TS	PG	VA	FC	CPM
B5	32S	0R	20S	30S	0R	22S	20S	26S	0R	24S	0R	18IR	20S
B9	20S	2R	30S	20S	0R	26S	26S	20S	0R	26S	0R	16IR	24S
B10	24S	22S	0R	36S	0R	42S	20S	0R	0R	20S	20S	24S	0R
B13	20S	0R	24S	24S	0R	20S	28S	20S	0R	40S	0R	18IR	22S
B17	28S	22S	8R	40S	8R	50S	28S	16IR	0R	22S	20S	28S	30S
B22	26S	20S	6R	28S	4R	40S	28S	4R	0R	30S	20S	22S	20S
B27	20S	2R	20S	22S	0R	26S	26S	20S	0R	28S	0R	6R	24S
B33	40S	12R	22S	26S	4R	24S	28S	0R	10R	28S	0R	16IR	30S
B35	30S	24S	10R	20S	4R	30S	24S	24S	0R	30S	30S	28S	20S

Penicillin: PG (10 µg), Oxacillin : Ox (1 µg), Rifampicin : RP (5 µg), Kanamycin : K (30 µg), Ciprofloxacin : CIP (5 µg), Imipenem: IMI (10 µg), Vancomycin : VA (30 µg), Amikacin : AK (30 µg), Linezolide : LZD (30 µg), Clindamycin : CD (2 µg), Sulfamethoxazole : TS (23.75 µg), Fusidic acid: FC (10 µg), Cefepime : CPM (30 µg) Resistant (R) ≤14 mm; intermediate resistant (IR) 15–19 mm; susceptible (S) > 19 mm

3.4. LAB bio-preservation effect on tomato

Figure 4 (a, b, c) showed the appearance of tomatoes after bio-preservation through immersion in supernatants and coating with cells for 2 weeks. Overall, the tomato surfaces treated with supernatant and cells remained intact and appeared smoother compared to the control (untreated), where the tomato surface was crumpled and the tomato showed signs of atrophy (Figure 4a and 4b). After 2 weeks of bio-preservation with both supernatant and cells, the tomato surface treated with supernatant was smoother than the surface of tomatoes treated with cells. In contrast, the untreated (control) tomatoes were crumpled and atrophied.

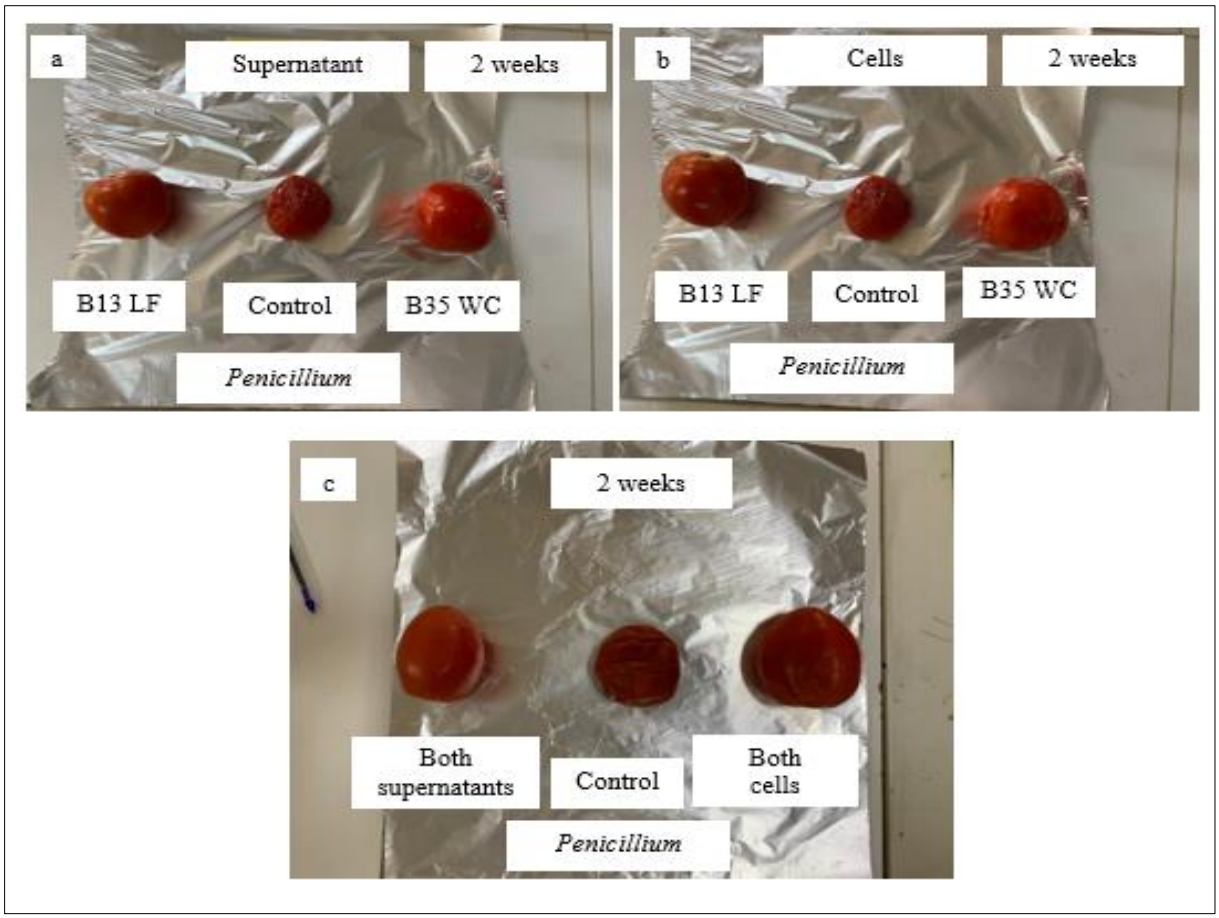

Figure 4 Tomato biopreservation by immersion and coating with supernatant and cells of *Limosilactobacillus fermentum* B13 and *Weissella confusa* B35

4. Discussion

Currently, most food contamination is caused by moulds. The fight against these spoilage agents in the agri-food sector, as well as the development of new strategies such as bio-preservation, is a promising approach [20-22]. Molecular identification of the selected lactic acid bacterial isolates revealed that they belonged to *Limosilactobacillus fermentum* and *Weissella confusa*. These species are commonly associated with the fermentation of various foods. Moreover, studies by Baek et al. [23] demonstrated that *Weissella confusa* and *Lactobacillus fermentum* exhibited antifungal activity against certain mould species such as *Cladosporium* sp., *Penicillium crustosum*, and *Neurospora* sp. This suggests that these species could be particularly useful in food bio-preservation. In this study, from 150 lactic acid bacteria strains previously tested (results not shown), only 9 strains exhibited inhibitory effects on all 5 phytopathogenic mould strains. This finding is consistent with the study by Guo et al. [24], which investigated the antifungal activity of 220 *Lactobacillus* strains isolated from various sources (cow, cheese, and cereals) against three dermatophytic fungi: *Microsporum canis*, *Microsporum gypseum*, and *Epidermophyton floccosum*. The results showed that only 8 strains demonstrated strong antifungal activity.

The antifungal activity of *Lactobacillus* strains has been elucidated in numerous studies. Abouloifa et al. [25] characterized the antifungal activity of 14 probiotic strains of *Lactobacillus* (*L. plantarum*, *L. pentosus*, and *L. brevis*) isolated from traditionally fermented green olives. They found that all the *Lactobacillus* strains and their cell-free supernatants exhibited strong antifungal activity against moulds (*Aspergillus niger*, *Penicillium* sp., *Fusarium oxysporum*, *Rhizopus* sp.), including inhibition of biomass and mycelium growth, as well as against yeasts (*Candida pelliculosa* and *Rhodotorula* sp.). In a similar approach, this study revealed that *Limosilactobacillus fermentum* exhibited antifungal activity against *Fusarium* sp., *Colletotrichum gloeosporioides*, *Botryodiplodia theobromae*, *Aspergillus carbonarius*, and *Aspergillus niger*, helping to retard and minimize their growth, thereby ensuring bio-conservation. This activity has been attributed to lactic acid bacteria, which produce numerous antifungal metabolites, including primary and secondary metabolites derived from fermentation and various degradation pathways used by these bacteria. These include organic

acids (lactic acid, acetic acid, phenyllactic acid, caproic acid), carbon dioxide, hydrogen peroxide, diacetyl, ethanol, fatty acids, peptides, reuterin, bioactive antimycotics, bacteriocins, and lactones, all of which have the ability to inhibit fungal growth [26]. Furthermore, they can degrade mycotoxins such as ochratoxins, aflatoxins, and Fusarium toxins [27].

The primary metabolites of lactic acid bacteria that significantly affect fungi by inhibiting mycelial growth are organic acids. Lactic acid, the main metabolite of lactic acid bacteria, causes a reduction in pH, which inhibits many microorganisms [28]. The undissociated, more hydrophobic form of the acid spreads across the cell membrane and dissociates inside the cell, releasing H+ ions that acidify the cytoplasm [29]. In addition to the pH effect, the undissociated acid causes a drop in the electrochemical proton gradient, leading to bacteriolysis and, ultimately, the death of susceptible bacteria.

Antibiotic susceptibility of LAB strains is one of the decisive criteria for strain selection from a safety perspective. LAB are also known to exhibit resistance to several antibiotics. They can carry and, in some cases, transfer antibiotic resistance genes [30]. A significant proportion of these bacteria, found in fermented foods, are thought to derive their resistance through mutations. Many *Lactobacillus* species exhibit natural (intrinsic) resistance, such as resistance to vancomycin and teicoplanin [30-31]. Several acquired resistances can also be found, such as resistance to chloramphenicol (cat gene) in *L. reuteri* and *L. plantarum* strains, and the tetracycline resistance gene tet(M) in *L. casei*, *L. plantarum*, *L. reuteri*, *L. rhamnosus*, and *L. lactis* strains [30]. The 9 strains selected in the present study showed antibiotic sensitivity. Bacteria used in the food industry can carry antibiotic resistance genes, which may be transferred to pathogenic bacteria. Indeed, antibiotic resistance can result in the transfer of resistance genes to other non-pathogenic normal flora [32-33] and even to consumers [34], which could pose a serious health risk.

Based on the visual appearance, the bio-preservation test on tomatoes using *Limosilactobacillus fermentum* B13 and *Weissella confusa* B35 showed that the treated tomatoes had smoother surfaces compared to the control (untreated tomatoes), and the tomatoes did not exhibit spoilage. These results suggested that the lactic acid bacteria species from *attiéké* (*cassava semolina*) fermentation could be applied in vegetable bio-preservation. The use of lactic acid bacteria species in vegetable bio-preservation has been reported by several authors. Various *Lactobacillus* species have exhibited antifungal activity against *Penicillium brevicompactum*, *Penicillium crustosum*, *Penicillium solitum*, *Penicillium glabrum*, *Penicillium palitans*, *Mucor plumbeus*, and *Mucor circinelloides* [35]. Several metabolites have been identified as contributing to the antifungal activity of lactic acid bacteria, including organic acids, fatty acids, proteinaceous compounds, diacetyl, and hydrogen peroxide (Shi and [35]. In addition to these substances, other antifungal metabolites have been discovered. According to Yang et al. [36], *Lactobacillus plantarum* AF1 produces a novel antifungal compound, δ-dodecalactone, which has a potent inhibitory effect on moulds, including *Penicillium roqueforti* and *Aspergillus* strains. Wang et al. [37] documented the antifungal properties of 2-propenyl ester, a benzeneacetic acid produced by *Lactobacillus plantarum* IMAU10014, which strongly affects *Penicillium citrinum* and *Penicillium drechsleri*. Ryan et al. [38] found that the cell-free supernatants of *L. amylovorus* DSM 19280 contained two antifungal nucleosides: cytidine and 20-deoxycytidine

5. Conclusion

This study showed the potential of cassava ferment as source of beneficial microorganisms for food bio-preservation. It allowed the identification of nine LAB strains belonging to *Limosilactobacillus fermentum* and *Weissella confusa*. Additionally, these LAB especially the strains B10, B13, and B35 display a variety of potential antifungal activities against spoilage moulds such as *Fusarium sp.*, *Colletotrichum gloeosporioides*, *Botryodiplodia theobromae*, *Aspergillus carbonarius*, and *Aspergillus niger*. Furthermore, all LAB strains have a typical pattern of antibiotic susceptibility. In particular, the strains *Limosilactobacillus fermentum* B13 and *Weissella confusa* B35 exhibit good bio-preservation of tomatoes stored at room temperature over a 2-week period. The analysis of the biochemical, nutritional, and phytochemical properties of tomatoes after the 2-week bio-preservation period is now under investigation.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare no conflicts of interest.

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Data Availability Statement

All the datasets generated in the present study are included in the manuscript. The partial 16 S rDNA sequences of LAB strains were deposited in the NCBI GenBank. Accession numbers.

- https://www.ncbi.nlm.nih.gov/nucleotide/OR458563
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- https://www.ncbi.nlm.nih.gov/nucleotide/OR458570
- https://www.ncbi.nlm.nih.gov/nucleotide/OR458571

References

[1] L. Lycia, and Y.L. Simoud, The nutritional, antioxidant and therapeutic properties of watermelon (Citrulluslanatus) (2020). https://dspace.ummto.dz/handle/ummto/14167.

[2] WHO. The World Health Report 2002 - Reducing Risks, Promoting Healthy Life. Education for Health: Change in Learning & Practice 16 (2) (2003) 230–230.

[3] K.Y. Kamelan, A.C. Kouakou, K.D Kra, F.H. Tra Bi, H.A. Diallo, K. Tano, Isolation and molecular identification of fungi associated with the spoilage of farms fruit in Southern Côte d'Ivoire, African Journal of Microbiology Research 8 (34) (2014) 3171–77.

[4] D. Desbordes, Microbiological quality of fruit and vegetables: flora, spoilage, health risks, prevention. Bibliographic Research Report, Normal school of information sciences and libraries (2003) 47p.

[5] M. Baddi, N. Saadia, S. Alali, A. EL Hraiki, The economic and health impact of mycotoxins between now and the future, Moroccan Journal of Agronomic and Veterinary Sciences (2021). 9 (3).

[6] M. Barth, T.R. Hankinson, H. Zhuang, F. Breidt, Microbiological spoilage of fruits and vegetables. In Compendium of the Microbiological Spoilage of Foods and Beverages, edited by William H. Sperber and Michael P. Doyle, (2009) 135–83. New York, NY: Springer.

[7] O.O. Akinmusire, Fungal species associated with the spoilage of some edible fruits in Maiduguri Northern Eastern Nigeria, Advances in Environment and Biology 5 (1) (2021) 157-161.

[8] R. Lahlali, M. Boulif, A. Moinina, Phytosanitary practices of apple growers: The case of the Fès-Meknès region, Moroccan Journal of Agronomic and Veterinary Sciences 9(2) (2021) 151-157.

[9] V.P. Singh, Recent Approaches in Food Bio-Preservation - a Review. Open Veterinary Journal 8 (1) (2018) 104–11.

[10] H. Bekkouche, B. Haouhat, S. Laala, Evaluation of the activity of lactic acid bacteria in the bio-preservation of certain foods. Dissertation for the award of the Academic Master's Degree. (2021). Ibn Khaldoun University - Tiaret. Algeria.

[11] S. Solvej, R. Balti, A.R. Neves, Bioprotective mechanisms of lactic acid bacteria against fungal spoilage of food. Current Opinion in Biotechnology, Food Biotechnology • Plant Biotechnology 56 (2019) 138–46.

[12] O.N. Djossou, Post-harvest mycoflora in robusta coffee and the use of lactic acid bacteria to control mycotoxigenic moulds and ochratoxin A, Doctoral thesis, Population biology and ecology Aix-Marseille 3, Environmental Sciences doctoral school (Aix-en-Provence, France) (2011).

[13] A. Gálvez, A. Hikmate, L.L. Rosario, N.B. Omar. Bacteriocin-based strategies for food bio-preservation. International Journal of Food Microbiology, 20th International ICFMH Symposium on Food Micro 120 (1) (2006) 51–70.

[14] I. Kaoutar El, N.S. Senhaji, S. Zinebi, R. Zahli, I. Haoujar, N. Amajoud, J. Abrini, K. El Ouardy, Potential application of bacteriocin produced from lactic acid bacteria, 48 (3) (2020) 237–51.

[15] R.V. Rajanikar, B.H. Nataraj, H. Naithani, S.A. Ali, N.R. Panjagari, P.V. Behare, Phenyllactic acid: a green compound for food biopreservation. Food Control, (2021) 128 108184.

[16] N. Saitou, M. Nei. The Neighbor-Joining Method: A new method for reconstructing phylogenetic trees. Molecular Biology and Evolution 4 (4) (1987) 406–425.

[17] K. Sudhir, G. Stecher, M. Li, C. Knyaz, K. Tamura. MEGA X: molecular evolutionary genetics analysis across computing platforms. Molecular Biology and Evolution 35 (6) (2018) 1547–1549.

[18] M. Balouiri, S. Moulay, K.I. Saad, Methods for in vitro evaluating antimicrobial activity: A Review. Journal of Pharmaceutical Analysis 6 (2) (2016) 71–79.

[19] CLSI. CLSI Supplement M100S. Performance standards for antimicrobial susceptibility testing, 26th ed.; Clinical and Labor-atory Standards Institute: Wayne, PA, USA, 2016.

[20] J-D. Kim, Antifungal activity of lactic acid bacteria isolated from Kimchi Against Aspergillus Fumigatus. Mycobiology 33 (4) (2005) 210–214.

[21] F. Daliri, A.A. Aboagye, E. B-M Daliri. Inactivation of foodborne pathogens by lactic acid bacteria. Journal of Food Hygiene and Safety 35 (5) (2020) 419–29.

[22] V. Lubomír, P. Ačai, A. Medvedová, Application of competitive models in predicting the simultaneous growth of Staphylococcus aureus and lactic acid bacteria in milk. Food Control 87 (2018) 145–152.

[23] E. Baek, H. Kim, H. Choi, S. Yoon, J. Kim, Antifungal activity of Leuconostoc Citreum and *Weissella confusa* in rice cakes. Journal of Microbiology 50 (5) (2012) 842–848.

[24] J. Guo, B. Brosnan, A. Furey, E. Arendt, P. Murphy, A. Coffey, Antifungal activity of Lac-tobacillus against Microsporum Canis, Microsporum Gypseum and Epidermophyton Floccosum. Bioengineered 3 (2) (2012) 104–113.

[25] H. Abouloifa, S. Gaamouche, Y. Rokni, I. Hasnaoui, R. Bellaouchi, N. Ghabbour, S. Karboune, Antifungal activity of probiotic Lactobacillus strains isolated from natural fermented green olives and their application as food bio-preservative. Biological Control 152 (2021) 104450.

[26] S. Crowley, J. Mahony, D. van Sinderen, Current perspectives on antifungal lactic acid bacteria as natural bio-preservatives. Trends in Food Science & Technology 33 (2) (2013) 93–109.

[27] F.A. Sadiq, Y. Bowen, F. Tian, J. Zhao, H. Zhang, W Chen, Lactic acid bacteria as antifun-gal and anti-Mycotoxigenic agents: A comprehensive review. Comprehensive Reviews in Food Science and Food Safety 18 (5) (2019) 1403–36.

[28] J. Schnürer, J. Magnusson, Antifungal lactic acid bacteria as bio-preservatives. Trends in Food Science & Technology, Second International Symposium on Sourdough - From Fundamentals to Applications 16 (1) (2005) 70–78.

[29] J.C. Piard, M. Desmazeaud, Inhibiting factors produced by lactic acid bacteria. Oxygen metabolites and catabolism end-products. Le Lait 71 (5) (1991) 525–541.

[30] M.S. Ammor, A.B. Flórez, B. Mayo. Antibiotic resistance in non-Enterococcal lactic acid bacteria and bifidobacteria. Food Microbiology 24 (6) (2007) 559–570.

[31] M. Bernardeau, J.P. Vernoux, S. Henri-Dubernet, M. Guéguen, Safety assessment of dairy microorganisms: the Lactobacillus genus. International Journal of Food Microbiology 126 (3) (2008) 278–285.

[32] G.J. Stewart, C.D. Sinigalliano, Detection of horizontal gene transfer by natural transformation in native and introduced species of bacteria in marine and synthetic sediments. Applied and Environmental Microbiology 56 (6) (1990) 1818–1824.

[33] H. Kruse, H. Sørum. Transfer of multiple drug resistance plasmids between bacteria of diverse origins in natural microenvironments. Applied and Environmental Microbiology 60 (11) (1994) 4015–4021.

[34] A.J. Baylay, L.J.V. Piddock, M.A. Webber. Molecular mechanisms of antibiotic resistance – Part I. In bacterial resistance to antibiotics – From molecules to man, 2019, 1–26. John Wiley & Sons, Ltd.

[35] C. Shi, M. Maktabdar. Lactic acid bacteria as bio-preservation against spoilage moulds in dairy products – A Review. Frontiers in Microbiology 12 (2022).

[36] H. Yang, Y. Sun, R. Cai, Y. Chen, B. Gu. The impact of dietary fiber and probiotics in infectious diseases. Microbial Pathogenesis 14 (2020) 103931.

[37] H.K. Wang, Y.H. Yan, J.M. Wang, H.P. Zhang, W. Qi. Production and characterization of antifungal compounds produced by Lactobacillus Plantarum IMAU10014. Plos one 7 (1) (2012) e29452.

[38] A.M.R. Liam, E. Zannini, F.D. Bello, A. Pawlowska, P. Koehler, E.K. Arendt. Lactobacillus Amylovorus DSM 19280 as a novel food-grade antifungal agent for bakery products. International Journal of Food Microbiology 146 (3) (2011) 276–283.