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Effect of Temperature, PH, and Salt on Bacteriocin Activity Produced from Different Strains of Lactic Acid Bacteria (LAB)

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ABSTRACT

The present study was conducted to assess the effect of temperature, pH, and salt on bacteriocin activity produced from different strains of Lactic Acid Bacteria. On the basis of cultural, biochemical, physiological and morphological identification the Lactobacillus species were identified. Four species of Lactobacillus were isolated from homemade cheese, raw milk, dosa paste, sauce, and curd. Lactobacillus species isolated from the sampled food source include; Lactobacillus brevis, Lactobacillus plantarum , Lactobacillus fermentum, and Lactobacillus casei. The bacteriocins activity towards different levels of temperature, pH and salt was recorded. The results showed that the highest bacteriocin activity was observed at 60°C for 180 min and the lowest bacterial activity was observed at 121°C for 20 min. L.plantarum(P2) reported the maximum bacteriocin activity.The highest bacteriocins activity was reported at pH-6. Across different bacterial strains tested for pH stability, L. plantarum(P2) reported maximum bacteriocins activity. It was observed that bacteriocins produced from L. brevis reported highest salt tolerance. Across different bacterial strains tested for salt tolerence, L. brevis reported maximum bacteriocins activity. The bacteriocin produced from LAB in the current experiment show resistance to pH, heat, and different organic solvents, thus this bacteriocin opened the prospect for its utilization in preservation of food products.

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Keywords: LAB, bacteriocins, temperature stability, ph stability, salt tolerance.

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I. INTRODUCTION

Lactic Acid Bacteria (LAB) have been intensively used in food preservation and research across the globe has reported that LAB stimulates the nutrient level of different fermented foods and its derived products (Sameh, *et.al.,* 2016; Raman, *et.al.,* 2022). LAB improves the flavor, taste, texture, and shelf-life of different products (Korcari, *et.al.,* 2021). The LAB creates lactic acid, which makes the environment more acidic and decreases the number of pathogenic microorganisms. LAB has the ability to create organic acids, hydrogen peroxide, diacetyl, and bacteriocins which are some of the antibacterial substances. These substances are known to lessen food deterioration and the growth or proliferation of harmful bacteria. As a result, the food industry has begun to pay more attention to the utilisation of these naturally occurring chemicals as food bio-preservative agents, which currently offers a viable alternative to chemical food preservation, particularly for ready-to-use products. (Suskovic *et.al.,* 2010, Lappa, *et.al.,* 2022). Bacteriocins could also be used to prepare products that aren't being sufficiently thermally sterilized at the time of there production, as there is a risk of product becoming contaminated with pathogenic microorganisms like *Listeria monocytogenes*, which has been linked to numerous outbreaks around the world (O'Sullivan *et.al.,*2002).

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Bacteriocins are ribosomally-synthesized peptides or proteins with antimicrobial activity, produced by different groups of bacteria. Many *Lactobacillus sp.* produce bacteriocins with rather broad spectra of inhibition Several LAB bacteriocins offer potential applications in food preservation, and the use of bacteriocins in the food industry can help to reduce the addition of chemical preservatives as well as well as the heat intensity treatments, resulting in foods that are better maintained naturally and have stronger nutritive and organoleptic qualities. This can be an alternative to satisfy the increasing consumers demands for safe, fresh-tasting, ready-to-eat, minimally-processed foods and also to develop "novel" food products (e.g. less acidic, or with a lower salt content) (Alkena *et.al.,* 2016). The inhibitory spectrum of some bacteriocins also includes food spoilage and/or food-borne pathogenic microorganisms. In the past years, a lot of works have been aimed to the detection, purification and characterization of bacteriocins, as well as to their use in food preservation strategies. However its essential to report the tolerance of bacteriocins towards different physical and chemical factors of environment. Keeping in view the role of bacteriocins, the present study was conducted to assess the effect of temperature, pH, and salt on bacteriocin activity produced from different strains of Lactic Acid Bacteria.

II. MATERIAL AND METHODOLOGY

2.1. Isolation and Identification of LAB

Homemade cheese, raw milk, dosa paste, sauce, curd, was serially diluted and spread on MRS agar medium. Plates were incubated anaerobically for 24-48 hrs at 30° . When required for the isolation of specific bacteria and incubated aerobically and anaerobically at various temperatures. A total of 13 isolates of bacterial species, namely *Aeromonas, Bacillus, Clostridium, Escherichia, Klebsiella, Listeria, Salmonella, Streptococcus, Staphylococcus, Enterobacter, Enterococcus,* and *Lactobacillus* were isolated from the sampled dairy products. On the basis of cultural, biochemical, physiological and morphological identification the *Lactobacillus* species were identified. Four species of *Lactobacillus* species were identified they include; *L.plantarum*(P2), *L. brevis, L.plantarum(P1)* , *L. casei,* and *L. fermentum*. The morphological characteristics of the bacterial strain obtained from different food sources is given in Table-1

Table-1: Morphological characteristics of the bacterial strain obtained from different food sources.

Strain code	Cell's form	Type	Colour	Motility test	Gram staining
L. brevis	Rod shaped	Bacilli	Yellow	Non-motile	Gram positive
L.plantarum	Slender rods	Coccobacilli	Yellow	Non-Motile	Gram positive
L.fermentum	Rod Shaped	Cocci	Creamy White	Non-Motile	Gram positive
L. casei	Rod-shaped	Smooth	Opaque with pigment	Non-Motile	Gram positive

2.2. Production of Bacteriocin

The bacteriocin production from the isolated strains of LAB species (*L.plantarum* (P2), *L. brevis, L.plantarum(P1), L. casei,* and *L. fermentum*) was grown in MRS broth(Hi Media Lab, Pvt Ltd.India) with 1% incoculumn and it was maintained at optimum culture condictions for 48 hours.The cells wer removed from the growth medium afer incubation by centrifugation at 15000r/min for 15 minutes at 4°C. The obtained cell-free supernatant was adjusted to pH 6.5 using 1mol/L NaOH and it was used as crude bacteriocin.

2.3. Purification of Bacteriocin

Two methods were used for this purpose; Ammonium salt precipitation and ion-exchange chromatography (Yang *et.al.,* 2018).

2.3.1. Ammonium Salt Precipitation

Ammonium salt precipitation, various concentrations of Ammonium Sulphate (10, 20, 30, 40, 50 and 60%) was added to 10 ml of crude bacteriocin in different salt of test tubes, precipitate for 24 h at 4°C. Then the obtained mixture of bacteriocin was centrifuged at 10,000 rpm for 10 min and then the obtained precipitate was further resuspended in 25ml of 0.05m of Potassium Phosphate buffer (pH 7)(Yang *et.al.,* 1992). Further dialysis was followed in dialysis bags against 2 litres of the same buffer.

2.3.2. Ion-Exchange Chromatography

The dialyzate was used for purification by cation exchange column (DEAE cellulose column) and elution was performed by using a linear gradient from citrate phosphate buffer ranging from pH 2.6 to 7.0 (Macher *et.al.,* 1980). The bacteriocin titer was assessed (Todorov et.al., 2004) of eluted samples.

2.4. Effect of Temperature, pH, and Salt on Bacteriocin Activity

In order to assess the impact of temperature, the purified bacteriocin activity was tested by incubating it at different temperatures between 60°C to 121°C and the residual activity was tested after 20 minutes, 40minutes, 120 minutes, and 150 minutes. To examine the affect the pH , the purified bacteriocin activity was tested by incubating it at different pH between 02 to 12 with sterile 1 mol/NaOH or 1 mol/L HCL. Further the effect of salt on the purified bacteriocin activity was observed by taking 400μl of the purified bacteriocin, and it was incubated at 37 °C with NaCl concentrations ranging from 10 to 40%. (Merck, Germany). Well diffusion assay was used to measure the activity.

III. RESULTS AND DISCUSSION

Four species of *Lactobacillus* were isolated from homemade cheese, raw milk, dosa paste, sauce, and curd. *Lactobacillus species* isolated from the sampled food source include; *Lactobacillus brevis*, *Lactobacillus plantarum* , *Lactobacillus fermentum,* and *Lactobacillus casei*. Table-1 and Table-2 shows the morphological, physiological and biochemical attributes of *L. brevis, L. plantarum* , *L. fermentum*, and *L. casei* isolated from cheese, raw milk, dosa paste, sauce, and curd. *Lactobacillus brevis* is a facultative anaerobic, hetero-fermentative species that is commonly found in fermented foods, such as beer, wine, and vegetables. It is a Gram-positive, non-spore-forming, rod-shaped bacterium that can occur singly, in pairs, or in short chains. The cells typically measure 0.7-1.0 μm in width and 2.0-4.0 μm in length, with rounded ends. *Lactobacillus plantarum* is a Gram-positive, non-motile, non-spore-forming, micro-aerophilic, and mesophilic bacterium. The cells are straight rods with rounded ends, measuring 0.9-1.2 μm in width and 3.0-8.0 μm in length, and can occur singly, in pairs, or in short chains. Some strains of *L. plantarum_* can possess true catalase and manganese-containing pseudo-catalase activities, as well as nitrate- and hematin-dependent nitrite reductases. *Lactobacillus fermentum* is a Gram-positive, non-spore-forming, rod-shaped bacterium that is commonly found in fermented foods and the human gastrointestinal tract. The cells are typically 0.5-0.8 μm wide and 2.0-4.0 μm long, with rounded ends, and can occur singly, in pairs, or in short chains. *Lactobacillus casei* is a Gram-positive, non-spore-forming, rod-shaped bacterium that is widely used in the production of fermented dairy products and as a probiotic. The cells are typically 0.5-0.8 μm wide and 1.0-10.0 μm long, with rounded ends, and can occur singly, in pairs, or in short chains.

Table-2: Physiological and Biochemical characteristics of Isolated *Lactobacilli.*

Characteristics	Raw milk	Curd	Cheeses	Sauce	Dosa Batter
Species	*L. plantarum* P1	*L. plantarum* P2	*L. casai*	*L. brevis*	*L. fermentum*
Morphology	Cream colony Gram +ve Rod	Cream Colony Gram +ve Rod	Cream Colony Gram +ve Rod	White colony Gram +ve Rod	White colony Gram +ve Rod
15 °C only	+	+	+	+	-
45 °C only	-	-	-	-	-
15 and 45 °C only	+	+	+	+	+
Acid and gas from glucose	-	-	-	+	+
NH_3 from Arginine	-	-	-	+	+
Arabinose	-	-	+	+	+
Cellobiose	+	+	-	-	+
Mannitol	+	+	-	-	+
Mannose	+	+	+	-	+
Melebiose	+	+	-	-	+
Raffinose	+	+	+	-	+
Ribose	+	+	+	+	+
Salicin	+	+	+	-	+
Lactose	-	-	-	+	+
Rhamnose	-	-	-	-	-
Sorbitol	-	-	-	-	+
Xylose	-	-	-	+	+
Trehalose	-	-	-	-_	+

Due to the fact that all bacteriocin was created during the pre- and early exponential growth phases and reached a maximum level at late stationary phase, the test isolates synthesis of bacteriocin demonstrated secondary metabolic kinetics. According to certain accounts, bacteriocins are created at all times during the experimental growth phase rather than just in the late logarithmic or early stationary phases.The process of producing bacteriocin was optimised by taking into account many parameters, including carbon and nitrogen sources, pH, temperature, and salt content. The optimum parameter was then determined using an arbitrary unit. The addition of glucose (2.0%) increased the amount of bacteriocin that could be produced, while the addition of other carbon sources had no effect or had a negative impact on production. For the nitrogen source, the medium's maximum production was achieved by mixing Tryptone, yeast extract, and meat extract together. While extremely alkaline and acidic pH did not enable the formation of bacteriocin, maximum activity in composition medium was attained at initial pH ranging from 6-8, and the optimal temperature was 300C.The bacteriocin was created at its highest level under the ideal conditions, and once it had been purified, it could be employed right away as a bio-preservative. The manufacturing of bacteriocins can be optimised to produce them more cheaply, which could eliminate the need to add chemical preservatives altogether or at least limit their use.

Table-3: Effect of Temperature on the activity (AU/µl) of bacteriocin produced by the isolated strains of LAB species.

Temperature	*L. plantarum(P1)*		*L. plantarum(P2)*		*L. casei*		*L. brevis*		*L. fermentum*	
	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl
60°C for 150 min	11.0	110	15.0	150	11.0	110	13.0	130	10	100
80°C for 120 min	9.0	90	13.0	130	10.0	100	8.0	80	7.0	70
100°C for 40 min	8.0	80	12.0	120	5.0	50	7.0	70	0	0
1210°C for 20 min	8.0	80	12.0	120	0	0	0	0	0	0

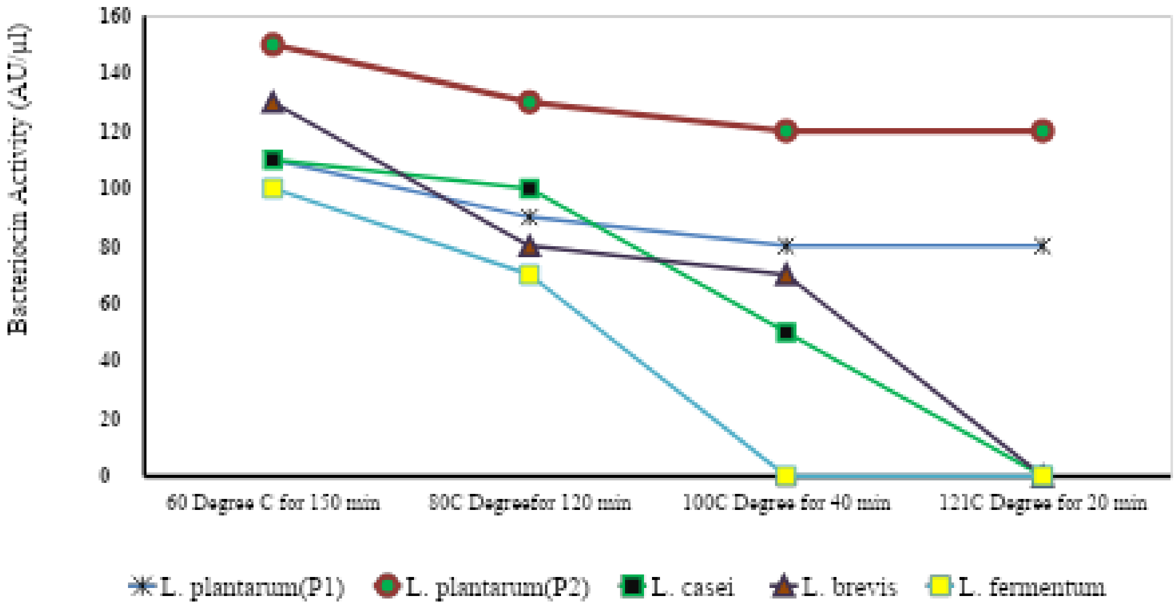

Fig-1: Thermal stability of Bacteriocin after exposure at different temperature treatment

The optimum condition for maximum bacteriocin activity was demonstrated by culturing the bacteria among various thermal ranges. Purified bacteriocin was exposed to various heat treatments: 60°C for 180 min, 80°C for 120 min, 100°C for 40 min and 121°C for 20 min (Table-3, Fig-1). The results showed that the highest bacteriocin activity was observed at 60°C for 180 min and the lowest bacterial activity was observed at 121°C for 20 min. Across different bacterial strains *L.plantarum*(P2) resported the maximum bacteriocin activity followed by *L. brevis, L.plantarum(P1) ,L. casei,* and *L. fermentum.* The results report that with the increase in the temperature there was a decrease in the activity (AU/µl) of bacteriocins. The *L.plantarum*(P2) showed the highest thermal stability that the other bacteriocins in terms of the temperature to which they were exposed.

Table 4: Effect of pH on the activity (AU/µl) of bacteriocin produced by the isolated strains of LAB species.

pH	*L. plantarum(P1)*		*L. plantarum(P2)*		*L. casei*		*L. brevis*		*L. fermentum*	
	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl
2	10	100	15	150	11	110	13	130	09	90
4	14	140	17	170	12	120	16	160	10	100
6	15	150	19	190	13	130	18	180	12	120
8	11	110	18	180	09	90	16	160	07	70
10	05	50	18	180	05	50	15	150	00	00
12	00	00	17	170	00	00	15	150	00	00

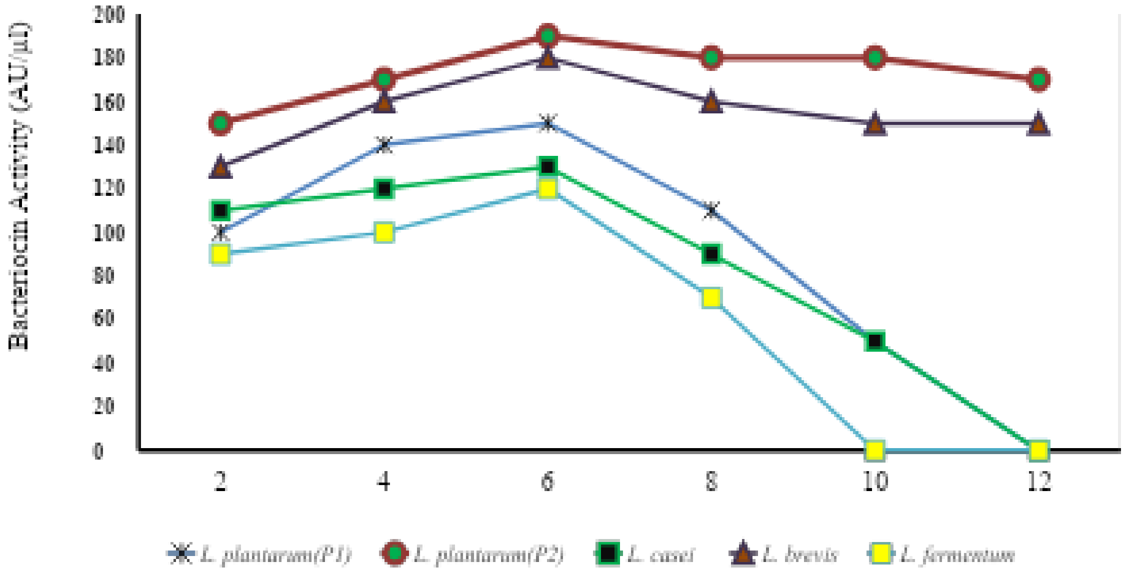

Fig-2: pH stability of Bacteriocins produced from different LAB species after exposure at different pH

The influence of pH on the activity of bacteriocins. Stability of pH of bacteriocins was observed at pH range of 2.0 to 12.0. It was observed that the bacteriocins activity report maximum stability between pH 4-6. The highest bacteriocins activity was reported at pH-6 (Table-4.18). Across different bacterial strains tested for pH stability, *L. plantarum*(P2) reported maximum bacteriocins activity followed by *L. brevis, L.plantarum(P1), L. casei,* and *L. fermentum* (Table-5, Fig-2). *L. plantarum* (P2) showed was reported to be the highest pH stable of bacteriocins after exposure at different pH. The overall results report that the pH affects the bacteriocins activity significantly.

Table-5: Effect of Salt on the activity (AU/µl) of bacteriocin produced by the isolated strains of LAB species.

Salt Conc.	*L. plantarum(P1)*		*L. plantarum(P2)*		*L. casei*		*L. brevis*		*L. fermentum*	
	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl	ZI (mm)	AU/µl
10%	12.0	120	19	190	11.0	110	20.0	200	11.0	110
20%	10.0	100	18	180	11.0	110	19.0	190	9.0	90
30%	8.0	80	16	160	7.0	70	18.0	180	8.0	80
40%	0.0	00	16	160	5.0	50	18.0	180	6.0	60

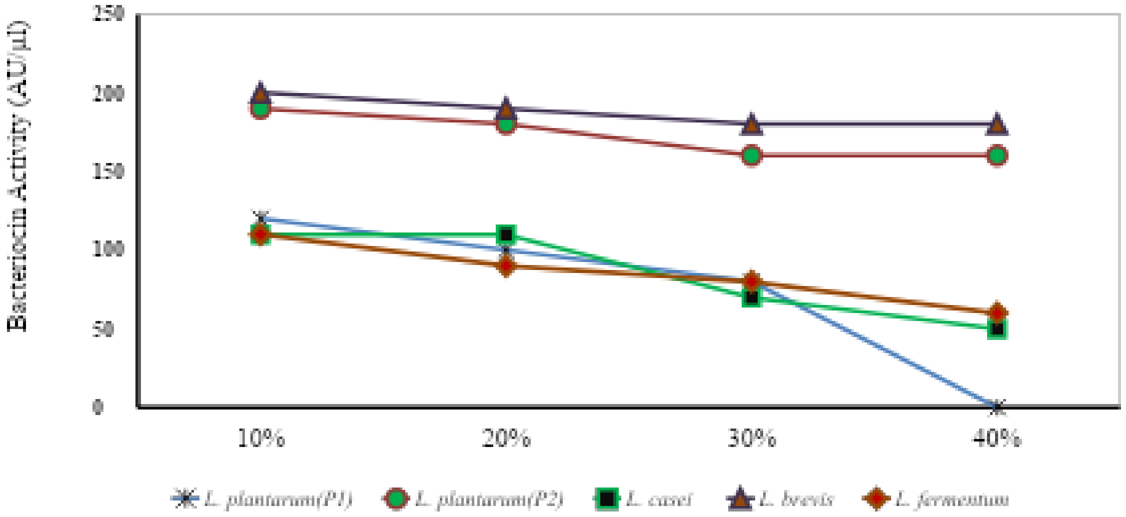

Fig. 3: Salt tolerance of Bacteriocins produced from different LAB species.

400µl of the purified bacteriocin were incubated at 37 °C with NaCl concentrations ranging from 10 to 40%. (Merck, Germany). Well diffusion assay was used to measure the activity. It was observed that bacteriocins produced from *L. brevis* reported highest salt tolerance. The tolerance by bacteriocin activity was recorded lowest by *L. casei*. Across different bacterial strains tested for salt tolerence, *L. brevis* reported maximum bacteriocins activity followed by *L. plantarum*(P2), *L.plantarum(P1), L. fermentum,* and *L. casei* (Table-5, Fig-3). The overall results show that the bacteriocin activity was highest at 20% salt concentration and lowest at 40% salt concentration.

IV. DISCUSSION

The present study examines the effect of temperature, pH and salt on the bacteriocin activity which were produced from *Lactobacillus species* isolated from Raw milk, Dosa, Curd, Sauces and Cheese. The *Lactobacillus species* were identified on the basis of morphological, biochemical and physiochemical attributes. Out of these five *Lactobacillus species*, two were identified as *L. plantarum* and coded as P1 and P2, respectively, due to their different potential in bacteriocin activity, and the other three were identified as *L. brevis, L. casai,* and *L. fermentum*. The bacteriocins produced by the *Lactobacillus species* in this investigation were subject to different levels of temperature, pH and salt to determine the stability of the bacteriocins for their future applications.

The *Lactobaccilus* bacteriocin isolated from different samples raw milk, dosa paste, curd, sauce and cheese have thermal stability at 60°C for 150 min. The results showed that the highest bacteriocin activity was observed at 60°C for 150 min and the lowest bacterial activity was observed at 121°C for 20 min. This is significant if the bacteriocin is to be utilized as a food preservative because many food preparation processes require a heating stage. The mechanism of heat-stability of LAB bacteriocins has been reported earlier for *Plantaricin S* (Jimenez-Diaz *et.al.,* 1990), *Plantaricin A* (Daeschel *et.al.,* 1985), *Plantaricin 149* (Kato *et.al.,* 1994), *Plantaricin SA6* (Rekhif *et.al.,* 1995), *Plantaricin 423* (Van-Reenen, 1998), *Plantaricin C19* (Audisio, 1999), *Pentocin TV35b* (Okkers *et.al.* ,1999), *L. brevis* 0G1 (Ogunbanwo *et.al.,* 2003), and *lactocin RN78* (Mojgani and Amirinia, 2007). Because we noted heat stability of *L. plantarum* P2 bacteriocin, the conclusions of the present study are in consistent with the findings of the papers cited above. This bacteriocin belongs to the category of bacteriocins with low molecular weight that are heat stable and retain their activity after heating at 121° C for 60 min. The bacteriocin is superior in processed foods when high heat is used because of this property. The thermal stability of other bacteriocins was not good. Additionally, Andersson (1986) noted a decrease in activity following a 15-minute heat treatment at 121° C.

The test isolates regarding bacteriocin's activity was also pH-dependent. The bacteriocin produced by *L. brevis, L.plantarum(P1), L.plantarum(P2), L. casei,* and *L. fermentum* showed pH stability at acidic pH (2-6). However only *L. brevis* and *L.plantarum(P2)* retained itheir activity at alkaline pH (8-12). *L. fermentum* have completely lost their activity form pH 10 and *L. brevis* lost its complete activity at pH 12 respectively. The overall bacteriocin activity was reported highest between pH 4-6. This was also demonstrated by Reddy *et.al.* (1984) and Abdel-Bar *et.al.* (1987) in their studies of two bacteriocins, bulgarican and lactobulgarican, isolated from *L. bulgaricus,* which had the highest activity and stability at pH 2.2 and 4.0, respectively, against a variety of pathogenic and spoilage bacteria.

In particular, pH 6 was found to have increased antibacterial activity in the bacteriocin generated by *L. brevis, L.plantarum(P1), L.plantarum(P2),1 L. casei,* and *L. fermentum*. This might be because bacteriocins' increased net charge at low pH makes it easier for the molecules to pass through the cell wall. At lower pH levels, bacteriocin solubility may also rise, aiding the diffusion of bacteriocin molecules. This concurred with earlier studies that demonstrated that the presence of NaCI enhanced the antimicrobial action of bacteriocins like nisin, leucocin F10, enterocin AS-48, and others. A modest quantity of NaCI also inhibited nisin action (Bouttefroy *et.al.,* 2000). Additionally, sodium chloride reduced the antilisterial efficacy of the antibiotics acidocin CH5 (at 1-2%; Chumchalova *et.al.,* 1998), lactocin 705, leucocins 4010 (at 2.5% NaCl; Hornbaek *et.al.,* 2004), pediocin, (at 6.5% NaCl; Jydegaard *et.al.,* 2000), curvacin (Vaerluyten). The ionic interactions between bacteriocin molecules and charged groups involved in bacteriocin binding to target cells may be interfered with by sodium chloride, which has a protective effect (Bhunia *et.al.,* 1988.) In addition, sodium chloride may cause bacteriocin structural alterations or modifications to the target cell's envelope (Lee *et.al.,* 1993) or changes in the cell envelope of the target organisms (Jydegaard *et.al.,* 2000).

The bacteriocin produced from LAB in the current experiment show resistance to pH, heat, and different organic solvents, thus this bacteriocin opened the prospect for its utilization in preservation of food products. It can also confirm that these LAB strains can be used as starting cultures for microorganisms and as bio-preservatives in a variety of fermented foods. The most typical bacteria for food fermentation and preservation are LAB. Their significance is primarily related to their safe metabolic activity as they consume accessible sugar while growing in food to produce organic acids and other metabolites (Gomashe *et.al.* 2014).

V. CONCLUSION

Lactic Acid Bacteria (LAB) can produce a range of antimicrobial compounds, such as organic acids, hydrogen peroxide, and bacteriocins, which can effectively inhibit the growth of various microorganisms. Four species of *Lactobacillus* were isolated from homemade cheese, raw milk, dosa paste, sauce, and curd. *Lactobacillus species* isolated from the sampled food source include; *Lactobacillus brevis*, *Lactobacillus plantarum* , *Lactobacillus fermentum,* and *Lactobacillus casei*. The bacteriocins activity towards different levels of temperature, pH and salt was recorded. The characterization of Bacteriocins from test isolates were studied in order to take advantage of their potential and make them viable candidates for deployment as a safe and effective biological preservative in the future. Across different LAB species the bacteriocins produced from *L. plantarum* report distinctive and comprehensive heat stability, pH instability, and salt tolerance, can be exploited as a biopreservative to increase the food items' hygienic and safety (especially processed foods).The use of Lactobacillus species as natural biopreservatives in food products is a promising approach to enhance food safety and extend shelf-life, thereby reducing the reliance on synthetic preservation methods.

Conflict of Interest

Author don't have any conflict of Interest regarding the publication of this manuscript

Plagiarism and AI Content

The author declares that Plagiarism of the manuscript is below 10% similarity level, and 0% AI content detected.

REFERENCES

1. Abdel-Bar, N., Harris, N.D. and Rill, R.L. (1987). Purification and properties of an antimicrobial substance produced by *Lactobacillus bulgaricus*. *J. Food Sci*. 52:411-415.
2. Alemka, A., Clyne, M., Shanahan, F., Tompkins, T., Corcionivoschi, N., & Bourke, B. (2010). Probiotic colonization of the adherent mucus layer of HT29MTXE12 cells attenuates Campylobacter jejuni virulence properties. *Infection and immunity*, *78*(6), 2812-2822.
3. Andersson, R.E., Daeschel, M.A. and Hassan, H.M. (1988). Antibacterial activity of plantaricin SIK-83, a bacteriocin produced by *Lactobacillus plantarum*. *Biochem*. 17:61-68.
4. Audisio, M.C., Oliver, G. and Paella, M.C.1999. Antagonistic effect of E.faecium J96 against human and poultry pathogenic Salmonellae species. *J. food Prot* 62: 751-755.
5. Bouttefroy, A., Linder, M., & Milliere, J. B. (2000). Predictive models of the combined effects of curvaticin 13, NaCl and pH on the behaviour of Listeria monocytogenes ATCC 15313 in broth. *Journal of Applied Microbiology*, *88*(6), 919-929.
6. Chumchalova, J., Stiles, J., Josephsen, J., & Plockova, M. (2004). Characterization and purification of acidocin CH5, a bacteriocin produced by Lactobacillus acidophilus CH5. *Journal of Applied Microbiology*, *96*(5), 1082-1089.
7. Daeschel, M.A., Mckenney, M.C. and Mcdonald (1985). Bacteriocidal activity of *Lactobacillus plantarum* C-11. *Food Microbiol*. 7:91-98.
8. Gomashe A.V. *et.al.,* (2014). Screening and Evaluation of antibacterial activity of bacteriocin producing lab against selected bacteria causing food spoilage. *International Journal of Current Microbiology and applied Sciences*, 3(8): 658-665.
9. Hornbaek, T., Brocklehurst, T. F., & Budde, B. B. (2004). The antilisterial effect of Leuconostoc carnosum 4010 and leucocins 4010 in the presence of sodium chloride and sodium nitrite

examined in a structured gelatin system. *International journal of food microbiology*, *92*(2), 129-140.
10. Jimenez-Diaz, R., Piard, J.C., Ruiz-Barba, J.L. and Desmazeaud, M.J. (1990). Isolation of a bacteriocin producing *Lactobacillus plantarum* strain from a green olive fermentation. Third symposium on lactic acid bacteria. *FEMS Microbiol Rev.* 87:91.
11. Jydegaard, A. M., Gravesen, A., & Knøchel, S. (2000). Growth condition‐related response of Listeria monocytogenes 412 to bacteriocin inactivation. *Letters in Applied Microbiology*, *31*(1), 68-72.
12. Kato, T.; Mastsuda, T.; Ogawa, E.; Ogawa, H.; Kato, H.; Doi, U.; Nakamura, R. (1994). *Plantaricin*-149, a bacteriocin produced by *Lactobacillus plantarum* NRIC 149. *J. Ferment.Bioeng*., 77: 277-282
13. Korcari, D., Secchiero, R., Laureati, M., Marti, A., Cardone, G., Rabitti, N. S., ... & Fortina, M. G. (2021). Technological properties, shelf life and consumer preference of spelt-based sourdough bread using novel, selected starter cultures. LWT, 151, 112097.
14. Lappa, I. K., Kachrimanidou, V., Alexandri, M., Papadaki, A., & Kopsahelis, N. (2022). Novel Probiotic/Bacterial Cellulose Biocatalyst for the Development of Functional Dairy Beverage. *Foods*, 11(17), 2586.
15. Lee, S., Iwata, T., Oyagi, H., Aoyagi, H., Ohno, M., Anzai, K., ... & Sugihara, G. (1993). Effect of salts on conformational change of basic amphipathic peptides from β-structure to α-helix in the presence of phospholipid liposomes and their channel-forming ability. *Biochimica et Biophysica Acta (BBA)-Biomembranes*, *1151*(1), 76-82.
16. Macher, B. A., & Klock, J. C. (1980). Isolation and chemical characterization of neutral glycosphingolipids of human neutrophils. *Journal of Biological Chemistry*, *255*(5), 2092-2096.
17. Mojgani N., Sabiri G., Ashtiani M. & Torshizi M. (2009). Characterization of Bacteriocins Produced by *Lactobacillus brevis* NM 24 and *L. fermentum* NM 332 Isolated from Green Olives in Iran. *The Internet Journal of Microbiology*. 51(2), 178-184.
18. Ogunbanwo S.T, Sanni, A.I. & Onilude A.A. (2003). Characterization of bacteriocin produced by *Lactobacillus plantarum* F1 and *Lactobacillus* brevis *OG1 African Journal of Biotechnology*, 2(8): 219-227.
19. Okkers, D.J., Dicks, L.M., Silvester, M., Joubert, J.J. and Odendaal, H.J. (1999). Characterization of pentocin TV35b, a bacteriocin like peptide isolated from *Lactobacillus pentosus* with a fungistatic effect on *Candida albicans*. *J. Appl. Microbiol.* 87:726-734.
20. O'Sullivan, L., Ross, R., and Hill, C. (2002). Potential of bacteriocin-producing lactic acid bacteria for improvements in food safety and quality. *Biochimie*, 84: 593–604.
21. Raman, J., Kim, J. S., Choi, K. R., Eun, H., Yang, D., Ko, Y. J., & Kim, S. J. (2022). Application of lactic acid bacteria (LAB) in sustainable agriculture: Advantages and limitations. *International Journal of Molecular Sciences*, 23(14), 7784.
22. Rekhif, N., Atrih, A., & Lefebvrexy, G. (1995). Activity of plantaricin SA6, a bacteriocin produced by Lactobacillus plantarum SA6 isolated from fermented sausage. *Journal of Applied Bacteriology*, *78*(4), 349-358.
23. Sameh, A. G., Rehab, M. A. E. B., Abo, B. F. A., & Gamal, F. M. G. (2016). In vitro evaluation of probiotic potential of five lactic acid bacteria and their antimicrobial activity against some enteric and food-borne pathogens. *African Journal of Microbiology Research*, 10(12), 400-409.
24. Suskovic, J., Kos, B., Beganovic, J., Pavunc, A.L., Habijanic, K., and Matosc, S. (2010). Antimicobial activity-the most improvement property of probiotic amd starter lactic acid bacteria. *Food Technol. Biotehnol*, 48:297-307.
25. Todorov, S. D., van Reenen, C. A., & Dicks, L. M. T. (2004). Optimization of bacteriocin production by Lactobacillus plantarum ST13BR, a strain isolated from barley beer. *The Journal of general and applied microbiology*, *50*(3), 149-157.

26. Van Reenen C.A., Dicks, L.M., and Chikindas M.L. (1998), Isolation, purification and partial characterization of plantaricin 423, a bacteriocin produced by *Lactobacillus plantarum*. *J. Appl. Microbiol.* 84:1131-1137
27. Yang, E., Fan, L., Yan, J., Jiang, Y., Doucette, C., Fillmore, S., & Walker, B. (2018). Influence of culture media, pH and temperature on growth and bacteriocin production of bacteriocinogenic lactic acid bacteria. *Amb Express,* 8(1), 1-14.
28. Yang, Y. C., Baker, J. A., & Ward, J. R. (1992). Decontamination of chemical warfare agents. *Chemical Reviews*, *92*(8), 1729-1743.