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Characterization, Acidifying, and Antibacterial Activity of Lactic Acid Bacteria Against Spoilage Strains Present in Chicken Meat

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ABSTRACT

Nowadays, through their beneficial roles, lactic acid bacteria have been used as natural additives for the formulation of several products especially in the food industry; as they reduce the pathogen load and improve the quality of fermented food. This work focuses on the technological study of five homolactic strains isolated from three fermented barley preparations belonging to three species: Lactobacillus plantarum, Pediococcus spp, and Lactococcus lactis ssp. The experiment focused on testing their acidifying activity and their antibacterial action against a range of spoilage strains isolated from chicken meat. The identification was carried out based on the study of the cultural, morphological characters and biochemical tests which revealed the presence of five strains namely: Escherichia Coli, Salmonella sp, Staphylococcus aureus, Proteus vulgaris, and Klebsiella pneumonia. The results obtained showed that the strains (SC1, SC4, and SC5) have acidifying activities, in parallel with a significant antibacterial effect compared to the other strains (SC2, SC3) whose respective values were 1.63±0.07%, 0.99±0.23% and 1.29±0.65% for SC1, SC4 and SC5. Moreover, the cell-free culture supernatant (CFCS) inhibition zones range from 19.5 ± 0.70 mm to 10.25 ± 0.35 mm.

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1. INTRODUCTION

In recent years, several studies have focused on the mechanism of action of microorganisms on the biotransformation of food and their potential application in the food industry and biotechnology sector (Wang *et al.,* 2021; Dimassi *et al.,* 2020; Rokni *et al.,* 2017). Lactic acid bacteria are beneficial microorganisms widely used in the fermentation of foods by participating in the improvement of their quality or the development of certain organoleptic characteristics (Hamdaoui *et al.,* 2023; Rodriguez *et al.,* 2009), reducing the growth of undesirable microorganisms and prolonging the storage phase (Delves-Broughton, 1990), the sugar fermentation process of lactic acid bacteria is divided into two metabolic pathways **Figure 1**.

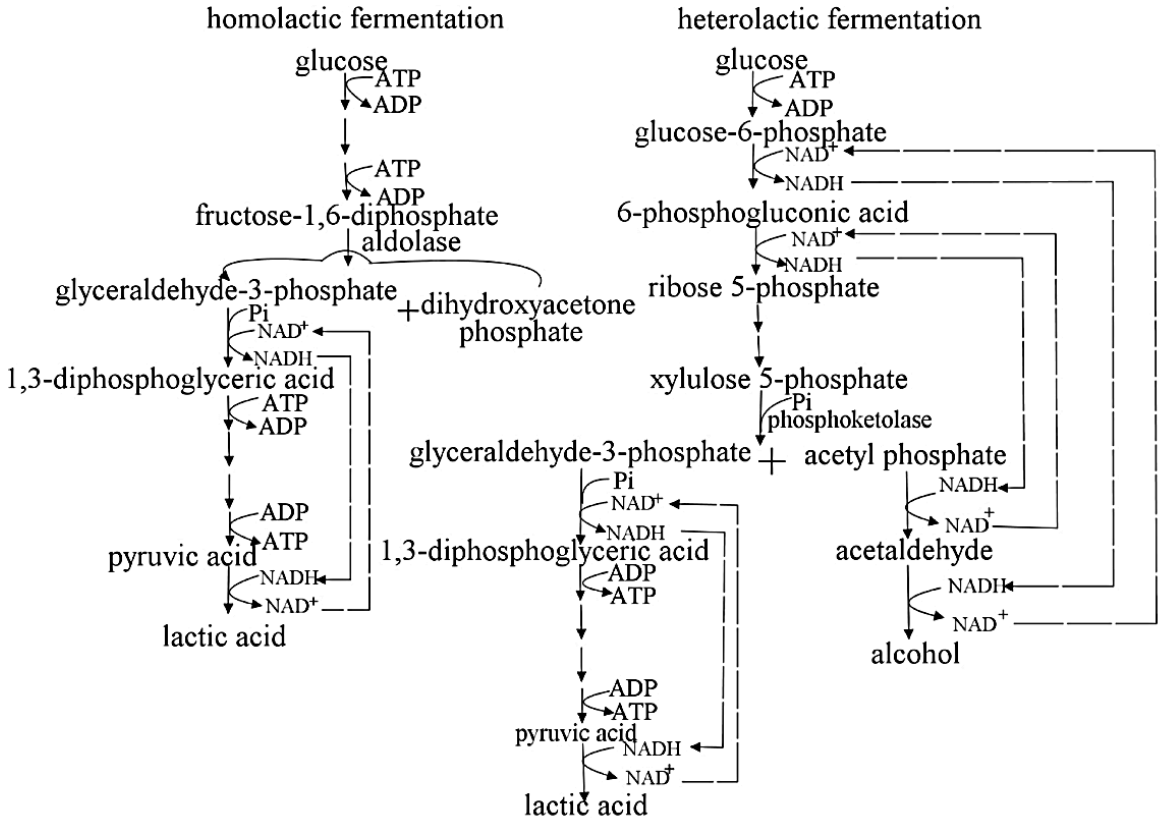

Figure 1. Different pathways of carbohydrate metabolic (Eiteman *et al.,* 2015).

There are many reports on bacteria (Rahayu, 2019; Sambegoro *et al*., 2021; Sari *et al*., 2023; Magno *et al*., 2022; Sari *et al*., 2022; Hamdaoui *et al*., 2023). Here, the main aim of this study was to select lactic strains with potential technological interest. For this reason, strain selection was based on several criteria. First, the strains are exposed to various acid, salt, and thermal stress conditions. Tolerance to these stress factors is an essential step in selecting potential candidates for application as zootechnical additives in poultry feed. To achieve this objective, the selection of strains was based on the evaluation of their acidifying power by measuring pH and acidity. The spoilage strains tested for antibacterial activity were isolated from chicken meat and identified by morphological and biochemical tests. The antibacterial effect was then verified using the well diffusion method (Barfoo *et al.,* 1983).

2. METHODS

2.1. Isolation and identification of lactic strains

The strains are isolated from three different barley-based biotopes on solid medium Man Rogosa Sharpes (MRS), purification is assured after a successive series of subculturing until macroscopically and microscopically distinct colonies are obtained. A pre-identification at the genus stage was carried out based on morphological, physiological, and biochemical tests and by the variation in conditions of NaCl, pH, and temperature on the growth of strains. The identification of the strains is then completed at the species stage by using an API 50CHL Medium gallery (Bio-Merieux reference 50410) containing 49 different fermentation profil, the species is given automatically thanks to a numerical profile by the computer-aided database of "APIWEB" version (5.1).

2.2. Acidifying activity in liquid MRS medium

The acidifying activity is evaluated by measuring the pH and determining the total acidity produced by the different cultures. Therefore, the cultures are inoculated into tubes containing 10 ml of liquid MRS medium and are incubated for two days (48 hours) at 30°C and in the dark (El Moualdi *et al.,* 2008). The pH is measured with an Orion Research pH meter with combined electrodes. Acidity is titrated with 0.1N NaOH using phenolphthalein as an indicator, and it is expressed as a percentage (%) of lactic acid (Ouhssine *et al.,* 2007) (see Equation [1]). The pH value and the acidity assay are taken relative to an uninoculated control.

$$\%\ lactic\ acid\ = \frac{Vol\ (NaOH).\ N\ (NaOH).\ \ Lactic\ acid\ weight\ (90{,}08)\ \ *100}{1000.\ (Sample\ mass)} \qquad (1)$$

2.3. Antibacterial activity

2.3.1. Screening of spoilage strains

2.3.1.1. Sample

The samples of chicken meat were purchased in the market from small local butchers from different areas of the city of Kénitra. After the slaughter of the animal, the samples were taken randomly from the chicken carcasses; the samples were collected aseptically and put in sterile plastic food bags bearing the date and the area from which the sample was taken. The samples were transported in a cooler to maintain a low temperature (2-4°C). Upon arrival at the laboratory, they were stored in a freezer at a temperature of -20°C until analysis.

2.3.1.2. Preparation of dilutions

25 g of each sample was placed in an Erlenmeyer flask containing 225 ml of sterile physiological water. A series of decimal dilutions to 10^{-6} was prepared from this stock solution.

2.3.1.3. Preparation of dilutions Isolation and identification of spoilage strains

Strains were isolated from broiler meat on selective media, and purity was ensured after several sub-cultures (Mourad & Nour-Eddine, 2006). Identification of the selected strains is carried out based on a study of cultural characteristics founded on growth conditions, thus the culture media for each strain are given in **Table 1**, followed by a morphological study based on macroscopic and microscopic observation of colonies, and biochemical tests for

catalase, cytochrome oxidase, nitrate reductase, motility, glucose and lactose fermentation with or without gas production (CO_2 and H_2S), citrate utilization, respiratory modes and amino acid metabolism. Coagulase and DNAase tests are reserved for the identification of staphylococci (Al-Joda & Jasim, 2021).

Table 1. Incubation conditions for the different spoilage strains.

Strain	Growth medium	Temperature(°C)	Time (hour)
Escherichia coli	Eosin Methylene Blue (EMB)	44	24
Proteus vulgaris	MacConkey	37	24-48
Staphylococcus aureus	Chapman	37	24
Klebsiella pneumonia	MacConkey	37	24-48
Salmonella sp	- Pre-enriched in buffered peptone Water	37 44	24 24
	-Enriched in Selenite-cysteine -Isolated in Salmonella-Shigella (SS) medium	37	24

2.4. Study of bacterial interaction

The Study of bacterial interaction is carried out using the agar diffusion well method, the lactic strains are seeded in MRS broth and incubated at 30°C for 18 to 24 hours; the supernatant is recovered after centrifugation at 10000 rpm for 10 min (Gagnon *et al.,* 2020). For pathogenic strains, they are cultivated on a solid medium Muller Hinton Agar (MHA) and incubated at 37°C for 18 to 24 hours.

The inoculum is prepared after inoculation of the indicator strain in sterile saline solution adjusted to a concentration of 10^8 cfu/ml to obtain a bacterial suspension with turbidity equal to 0.5 McFarland standards (Valgas *et al.,* 2007). The inoculum is swabbed into Petri dishes containing Muller-Hinton Agar medium with 5 mm diameter, wells which are then filled with 80 µl of cell-free culture supernatant (CFCS) previously recovered from the lactic acid strain (Gagnon *et al.,* 2020). The results were read after 24 hours of incubation at 35°C ±2. Each test was repeated three times at different times to minimize experimental error.

2.5. Statistical analysis

Each experiment was carried out in triplicate. Data collected were expressed as means ± standard deviation. One-way analysis of variance was used to determine significant differences in pH and acidity measurements between bacterial strains. In addition, a correlation test was performed to determine the relationship between the two factors. The results were analyzed using SPSS version 23 software and the levels of significance were established at a threshold of $p < 0.01$.

3. RESULTS AND DISCUSSION

3.1. Origin of lactic strains studied

The five strains studied were previously isolated and identified in the laboratory. The results of the pre-identification tests showed that the strains are gram-positive, immobile, in different forms: coccobacillus (SC1), bacillus (SC2), and cocci (SC3, SC4, SC5), and that they do not possess nitrate reductase or catalase. They are anaerobic but aerotolerant of homofermentative type and are resistant to various stress conditions as described in **Table 2**.

The ability to ferment various sugars was variable from one species to another and it was performed using the API 50CHL Medium gallery (Bio-Merieux reference 50410), the three species found are shown in **Table 3**.

Table 2. The ability to tolerate different stress conditions.

Lactic strain	pH			NaCl (%)			Temperature (°C)		
	4	5	6	0,5	1	1,5	18	20	22
SC1	+	+	+	+	+	+	+	+	+
SC2	-	+	+	+	+	+	+	+	+
SC3	-	+	+	+	+	+	-	+	+
SC4	+	+	+	+	+	+	+	+	+
SC5	+	+	+	+	+	+	+	+	+

(-) No growth, (+) Growth.

Table 3. Origin of lactic strains.

Codes	Species	ID	Biotope
SC1	*Lactobacillus plantarum*	99.8%	10g barley + 90ml traditional fermented milk
SC2	*Lactobacillus plantarum*	99.4%	10g barley + 90ml traditional fermented milk
SC3	*Lactococcus lactis ssp*	82.7%	10g barley + 90ml raw milk
SC4	*Pediococcus spp*	87.8%	10g barley + 90ml raw milk
SC5	*Lactococcus lactis ssp*	98.2%	10g barley + 90ml MRS medium

ID: the reliability of the identification given by apiweb™

3.2. Acidifying activity

Initially, the study focused on testing the acidifying activity of five strains of lactic acid bacteria in pure culture. The results illustrated in **Figure 2** revealed that the best measurements of acidifying activity are recorded on the three strains SC1, SC4, and SC5; whose pH values are 3.49±0.65, 3.96±0.05 and 3.64±0.07 with an acidity degree of 1.63±0.07%, 0.99±0.23% and 1.29±0.65% respectively. While the two strains SC2 and SC3 show less acidifying activity insofar as it is manifested by a pH of 4.39 ± 0.08 and 4.56 ± 0.05 with an acidity of 0.90 ± 0.04% and 0.77 ± 0.45%. The acidifying property is diverse not only between the different lactic strains but also between strains of the same species. These results are in line with those reported by Meghoufel: three strains of *lactococcus lactis* isolated from Jben showed variations in pH and acidity values. After 24 hours of incubation, the measured pH values were 4.05, 5.19, and 5.19, with Dornic densities reaching 124, 36, and 53 degrees Dornic, respectively. Studies conducted by Ouhssine *et al*. also highlighted differences in pH and acidity between different strains of *Lactococcus* and *Lactobacillus* after two days of incubation in the MRS broth. The measured pH values for four strains of Lactobacillus isolated from molasses and cow's milk were 4.1, 3.8, 3.7, and 4.3, with acidity degrees of 0.94%, 1.20%, 1.23%, and 1.02%, respectively. Furthermore, three strains of *Lactococcus* isolated from molasses, cow's milk, and strawberry juice presented pH values of 4.0, 3.5, and 4.2, with respective acidity degrees of 0.90, 1.61, and 0.70 (Ouhssine *et al.*, 2007).

The results of the correlation analysis show a strong negative correlation between the final pH and the acidity (%), meaning that if the acidity is high, the final pH will be low. This correlation is statistically significant at a 0.01 level of significance (**Table 4**).

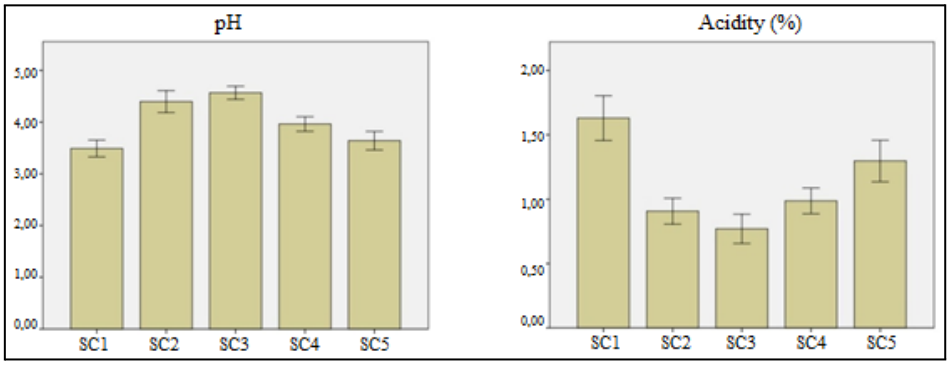

Figure 2. Acidity and pH of five lactic strains cultured in MRS broth during two days of incubation at 30°C.

Table 4. The correlation between pH and acidity.

		Final pH	Acidity (%)
Final pH	Pearson correlation	1	-,928**
	Sig (two-sided)		,000
	N	15	15
Acidity (%)	Pearson correlation	-,928**	1
	Sig (two-sided)	,000	
	N	15	15

(**) Correlation is significant at the 0.01 level (two-tailed).

3.3. Antibacterial activity

The characterization of the lactic strains from the technological point of view is completed by verifying their antibacterial effect against five spoilage strains isolated from chicken meat which are then identified through a biochemical gallery.

3.3.1. Screening of spoilage bacteria on chicken meat

3.3.1.1 Morphological test

The morphological test results shown in **Figure 3** revealed that the colonies' appearances are in different shapes, colors, and sizes. Also, these strains are gram-negative in bacilli form except for the SP3 strain which is gram-positive and in cocci form. Some are motionless like SP3 and SP5.

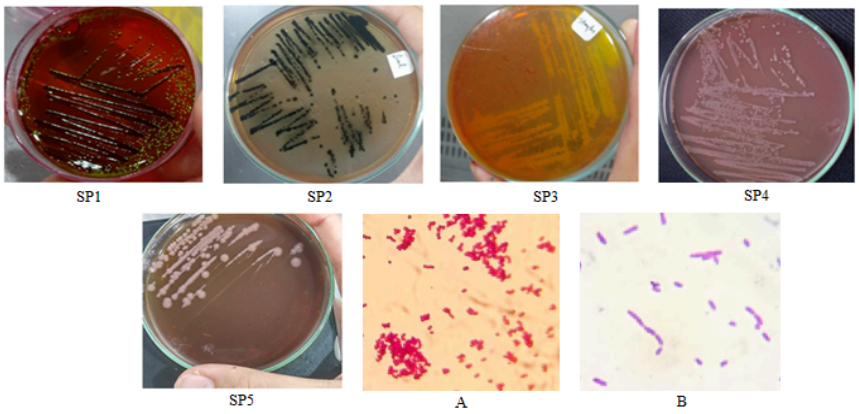

Figure 3. Macroscopic and microscopic appearance of strains. (SP1) *Escherichia Coli*, (SP2) *Salmonella sp*, (SP3) *Staphylococcus aureus*, (SP4) *Proteus vulgaris*, (SP5) *Klebsiella pneumonia*, (A) cocci form, (B) bacilli form.

3.3.1.2. Biochemical test

3.3.1.2.1. Energy metabolism

Subsequently, the identification is completed by various biochemical tests. The results showed that the five isolated strains have the catalase and nitrate reductase enzymes. Likewise, they are cytochrome oxidase negative and develop in anaerobiosis.

3.3.1.2.2. Protein metabolism

On the other hand, the strains SP1 and SP4 produce the indole from the degradation of the amino acid tryptophan, the presence of the enzymes: phenylalanine deaminase (PDA), tryptophan deaminase (TDA), lysine decarboxylase (LDC), arginine dihydrolase (ADH), ornithine decarboxylase (ODC), phenylalanine malonate and urease depend on the strain studied (**Table 5**). Furthermore, the strain SP3 has a positive deoxyribonuclease (DNase) and coagulase.

Table 5. Amino acid metabolism of spoilage strains.

Strain studied	SP1	SP2	SP3	SP4	SP5
PDA	-	-	Nt	+	-
TDA	-	-	Nt	+	-
LDC	+	+	-	-	+
ADH	-	+	+	-	-
ODC	+	+	-	-	-
Phenylalanine malonate	-	-	Nt	+	-
Urease	-	-	+	+	+
DNase	Nt	Nt	+	Nt	Nt
Coagulase	Nt	Nt	+	Nt	Nt

(-) Negative reaction, (+) Positive reaction, (Nt) Non-tested.

3.3.1.2.3. Carbohydrate metabolism

It is also noted that all strains are capable of fermenting glucose and mannitol, while the fermentation of lactose, malonate, saccharose, melibiose, and the formation of H_2S are variable from one species to another. However, all strains use citrate as a carbon source and produce CO_2 gas except for the SP1 strain. The test identification revealed the following strains: *Escherichia Coli* (SP1), *Salmonella sp* (SP2), *Staphylococcus aureus (*SP3), *Proteus*

vulgaris (SP4), and *Klebsiella pneumonia* (SP5). The result is similar to the one described in the clinical bacteriology manual by Avril *et al*. and Cowan and Steel's manual for the identification of medical bacteria. According to the identification protocol proposed by Barrow and Feltham, the strains SP1, SP2, SP4, and SP5 may be classified as *Enterobacteriaceae* species, in contrast, the SP3 strain is related to the family *Staphylococcaceae*.

These species are among the bacteria responsible for meat contamination. Some researchers have found that their growth is favored by several factors; Wardhana *et al*. reported that among 60 samples of chicken meat collected from local markets in Surabaya presents a high microbial contamination. The meat samples were contaminated with *Staphylococcus aureus* 58.3%, *Salmonella sp* 48.3% and *Escherichia Coli* 40%; due to lack of hygiene and poor quality of water used during handling (Wardhana *et al.,* 2021). Balcha and Gebretinsae reported that the way of handling the meat, the surface of exposure to the air, and the equipment used also contribute to the contamination, since the enumeration of total aerobic germs is higher from the table and the knife of 6.56 log cfu/cm^2 and 6.78 log cfu/cm^2, respectively. It is shown that 92% of the handlers in the Mekelle butcher shops were handling coins and serving simultaneously; which explains the presence of *Escherichia Coli* in 32% of the meat and contact surface samples collected from the butcher shops (Endele & Hailay, 2013). Another study was conducted on a total of 50 samples of raw chicken meat randomly purchased from local butchers in the city of Mansoura, Egypt, the results demonstrate that different species of *enterobacteria* are common in meat. Thus, *Proteus spp* 78.0% was found to be the most abundant, followed by *Klebsiella spp* 26.0% and *Escherichia coli* 16.0%. Theocharidi *et al*. reported that 81.8% strains of *Klebsiella pneumonia* were found in 90 out of 110 meat samples (Theocharidi *et al.,* 2022). The contents of the digestive tract can also contaminate the meat carcass during evisceration; this leads to contamination due to *Enterobacteriaceae* (Fosse *et al.,* 2006). Furthermore, it has been shown that poultry facilities and animal bedding during the rearing phase are also considered a source of microbial contamination of poultry (Gomes *et al.,* 2022).

These food-borne pathogens represent a major public health problem worldwide despite the regulations and control measures in place, yet they remain the main source of infectious diseases in developing and least-developed countries (Atlabachew & Mamo, 2021). However, controlling sanitary requirements by applying good hygiene practices makes it possible to avoid contamination and microbial proliferation.

3.3.2. Inhibition spectrum

The study of the antibacterial activity is carried out by measuring the diameter of the inhibition zones which appear around the wells (Pulsani *et al.,* 1979). The diameter of the zone of inhibition greater than 1 mm is reported as positive (Schillinger *et al.,* 1989).

The results in **Table 6** show that all lactic strains can inhibit all of the spoilage strains tested. The SC1 strain shows strong inhibitory activity against *Escherichia coli* and *Klebsiella pneumonia* with an inhibition diameter of 16.5 ± 0.70mm. The same result is noted for the SC5 strain against *Escherichia coli, Salmonella sp,* and *Klebsiella pneumoniae.* The most pronounced inhibitory effect of strain SC4 was against *Klebsiella pneumoniae* with an inhibition diameter of 19.5 ± 0.70mm. The SC2 strain expressed its strong antibacterial effect against *Salmonella sp* with an inhibition zone of 15.5 ± 0.65mm. The best inhibition observed in strain SC3 was against *Escherichia coli,* with an inhibition diameter of 15 ± 0.50 mm. However, the least active inhibition reported was that of strain SC3 against *Staphylococcus aureus* with an inhibition zone of 10.25 ± 0.35 mm, while the strongest

inhibition was that of strain SC4 against *Klebsiella pneumoniae* with an inhibition diameter of 19.5 ± 0.70 mm. **Figure 4** provides this interpretation.

Table 6. Diameter of the zone of inhibition of five lactic acid bacteria against five spoilage strains by the well diffusion method on Muller Hilton medium after 24 hours of incubation at 35±2°C.

Strain	Inhibition diameter in mm				
	SC1	**SC2**	**SC3**	**SC4**	**SC5**
SP1	16,5 ± 0,70	13 ± 0,50	15 ± 0,50	15,5 ± 0,70	16,5 ± 0,61
SP2	15 ± 0,55	15,5±0,65	13±0,41	14 ± 0,70	16,5 ± 0,78
SP3	14 ± 0,66	12,75 ± 0,35	10,25 ± 0,35	14 ± 0,00	15 ± 0,70
SP4	14,25 ± 0,35	15 ± 0,70	10,5 ± 0,70	12,5 ± 0,68	16 ± 0,25
SP5	16,5 ± 0,73	14 ± 0,00	12,5 ± 0,70	19,5 ± 0,70	16,5 ± 0,70

Diameter of the inhibition zone including the well diameter of 5mm.

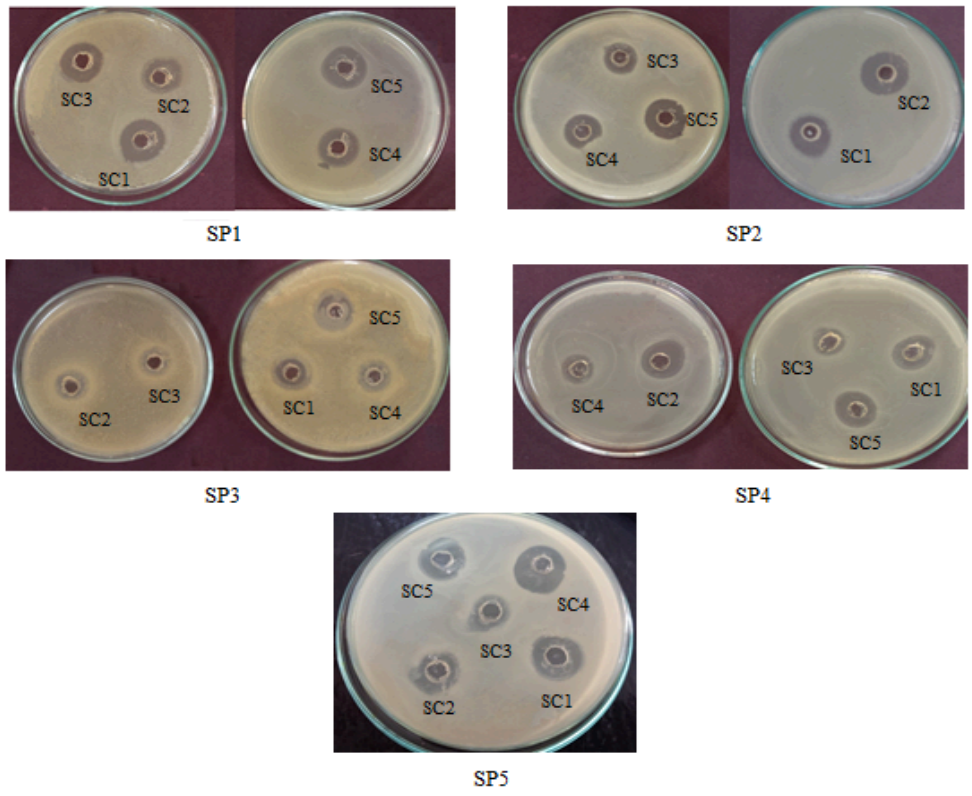

Figure 4. Inhibition obtained by five lactic strains (SC1, SC2, SC3, SC4, and SC5) against (SP1) *Escherichia Coli*, (SP2) *Salmonella sp,* (SP3) *Staphylococcus aureus,* (SP4) *Proteus vulgaris*, and (SP5) *Klebsiella pneumonia.*

In this context, Nizori A. *et al*, reported that lactic acid bacteria isolated from fermented durian flesh (tempoyak- traditional fermented food) exhibited a broad spectrum of inhibition against pathogenic and spoilage bacteria, with the supernatants of the strains effectively inhibiting the growth of *Staphylococcus aureus* and *Escherichia coli* ATCC 25923, with inhibition zone diameters ranging from 16.00 to 17.50 mm and 18.28 to 19.00 mm, respectively (Nizori *et al.,* 2019). Others have found that the antibacterial effect of cell-free supernatant (CFS) from seven strains of lactic acid bacteria isolated from traditional fermented dairy products possesses broad-spectrum antibacterial activity in vitro, against

food spoilage and pathogenic bacterial strains, with the zone of inhibition ranging from 5 to 16 mm in diameter (Girma & Aemiro, 2021). In brief, the antibacterial activity and acidifying power of lactic acid bacteria are often solicited in the preparation of fermented products (Kabrite *et al.,* 2019).

4. CONCLUSION

Lactic acid bacteria are widely used in the field of biotechnology thanks to their technological and functional as well as nutritional quality. The study of the acidifying activity showed that the SC1, SC4, and SC5 strains exhibit a strong acidifying power compared to the other SC2 and SC3 strains. This metabolic property is highly variable in lactic acid bacteria. In contrast, the identification of the alteration strains tested for the study of the antibacterial activity gave five species: Escherichia Coli, Salmonella sp, Staphylococcus aureus, Proteus vulgaris, and Klebsiella pneumonia. These species are often responsible for food toxin infection and foodborne illnesses. The SC1, SC4, and SC5 strains selected for their high acidifying activity also expressed a generally high antibacterial effect against the five spoilage strains tested. These two properties contribute to microbial safety and the enhancement of fermented products. The results of this study encourage possible use as a digestibility enhancer for poultry.

5. AUTHORS' NOTE

The authors declare that there is no conflict of interest regarding the publication of this article. Authors confirmed that the paper was free of plagiarism.

6. REFERENCES

AL-Joda, B. M. S., and Jasim, A. H. (2021). Biochemical testing revision for identification several kinds of bacteria. *Journal of University of Babylon for Pure and Applied Sciences*, *29*(2), 168-176.

Atlabachew, T., and Mamo, J. (2021). Microbiological quality of meat and swabs from contact surface in butcher shops in Debre Berhan, Ethiopia. *Journal of Food Quality, 2021*, 1-11.

Barefoot, S. F., and Klaenhammer, T. R. (1983). Detection and activity of lactacin B, a bacteriocin produced by Lactobacillus acidophilus. *Applied and Environmental Microbiology*, *45*(6), 1808-1815.

Delves-Broughton, J. (1990). Nisin and its uses as a food preservative. *Food Technology*, *44*, 100-117.

Dimassi, O., Iskandarani, Y., Afram, M., Akiki, R., and Rached, M. (2020). Production and physicochemical properties of labneh anbaris, a traditional fermented cheese like product, in Lebanon. *International Journal of Environment, Agriculture and Biotechnology*, *5*(3), 509-516.

Eiteman, M. A., and Ramalingam, S. (2015). Microbial production of lactic acid. *Biotechnology Letters*, *37*, 955-972.

El Moualdi, L., Labioui, H., Boushama, L., Benzakour, A., Ouhssine, M., and Yachioui, E. M. (2008). Activité bactéricide d'une souche de Lactococcus lactis subsp. cremoris. *Bull Soc Pharm Bordeaux*, *147*, 7-18.

Endale, B. G., and Hailay, G. (2013). Assessment of bacteriological quality of meat contact surfaces in selected butcher shops of Mekelle city, Ethiopia. *Journal of Environmental and Occupational Science, 2*(2), 61-66.

Fosse, J., Cappelier, J. M., Laroche, M., Fradin, N., Giraudet, K., and Magras, C. (2006). Viandes bovines: une analyse des dangers biologiques pour le consommateur appliquée à l'abattoir. *Rencontre Recherche Ruminants*, *13*, 411-414.

Gagnon, M., Ouamba, A. J., LaPointe, G., Chouinard, P. Y., and Roy, D. (2020). Prevalence and abundance of lactic acid bacteria in raw milk associated with forage types in dairy cow feeding. *Journal of Dairy Science*, *103*(7), 5931-5946.

Girma, A., and Aemiro, A. (2021). Antibacterial activity of lactic acid bacteria isolated from fermented Ethiopian traditional dairy products against food spoilage and pathogenic bacterial strains. *Journal of Food Quality, 2021*, 1-10.

Gomes, B., Pena, P., Cervantes, R., Dias, M., and Viegas, C. (2022). Microbial contamination of bedding material: One health in poultry production. *International Journal of Environmental Research and Public Health*, *19*(24), 16508.

Hamdaoui, N., Rokni, Y., Asehraou, A., Mouncif, M., Mennane, Z., Omari, A., Sellam, A., Hammouti, B., and Meziane, M. (2023) Technological aptitude and sensitivity of lactic acid bacteria leuconostoc isolated from raw milk of cows: From step-by-step experimental procedure to the results. *Indonesian Journal of Science and Technology 8*(2), 157-170.

Kabrite, S., Bou-Mitri, C., Fares, J. E. H., Hassan, H. F., and Boumosleh, J. M. (2019). Identification and dietary exposure assessment of tetracycline and penicillin residues in fluid milk, yogurt, and labneh: A cross-sectional study in Lebanon. *Veterinary World*, *12*(4), 527.

Magno, I. T., Esmail, R. P., Lim, M. S. S., Padua, J. N. J. D., Reyes, K. L., Tugom, L. A. G., Valdez, A. G., and Abusama, H. P. (2022). Antibacterial effect of calabash (crescentia cujete) leaf and fruit extract on preservation of lettuce (lactuca sativa) leaves with escherichia coli. *ASEAN Journal of Science and Engineering*, *2*(1), 91-94.

Mourad, K., and Nour-Eddine, K. (2006). Physicochemical and microbiological study of "shmen", a traditional butter made from camel milk in the Sahara (Algeria): isolation and identification of lactic acid bacteria and yeasts. *Grasas y Aceites*, *57*(2), 198-204.

Nizori, A., Sukendra, A., and Mursyid, S. (2019, November). Antimicrobial activity of lactic acid bacteria isolated from fermented durian flesh (tempoyak) against pathogenic and spoilage bacteria during storage. *IOP Conference Series: Earth and Environmental Science, 347*(1), 012053.

Ouhssine, M., and Elyachioui, M. (2007). L'application des déchets traités de l'algue Gelidium sesquipédale dans la culture du Maïs. *Afrique Science: Revue Internationale des Sciences et Technologie*, *3*(2), 259-270.

Pulsani, S. R., Rao, D. R., and Sunki, G.R. (1979). Antimicrobial Activity of lactic cultures: Partial purification and characterization of antimicrobial compounds produced by *Streptococcus thermophillus*. *Journal Food Sciences*, *44*, 575-578.

Rahayu, Y.S. (2019). Isolation and identification of hydrocarbon degradation bacteria and phosphate solubilizing bacteria in oil contaminated soil in Bojonegoro, East Java, Indonesia. *Indonesian Journal of Science and Technology*, *4*(1), 134-147.

Rodríguez, H., Curiel, J. A., Landete, J. M., de las Rivas, B., de Felipe, F. L., Gómez-Cordovés, C., and Muñoz, R. (2009). Food phenolics and lactic acid bacteria. *International Journal of Food Microbiology*, *132*(2-3), 79-90.

Rokni, Y., Abouloifa, H., Bellaouchi, R., Gaamouche, S., Mchiouer, K., Hasnaoui, I., Lamzira, Z., Ghabbour, N., and Asehraou, A. (2017). Technological process of fermented olive. *Arabian Journal of Chemical and Environmental Research*, *4*, 63-91

Sambegoro, P., Fitriyanti, M., Budiman, B. A., Kamarisima, K., Baliwangi, S. W. A., Alverian, C., Bagherzadeh, S., Narsimhan, G., Aditiawati, P., and Nurprasetio, I. P. (2021). Bacterial cell inactivation using a single-frequency batch-type ultrasound device. *Indonesian Journal of Science and Technology*, *6*(1), 65-80.

Sari, D. P., Yuniar, S., Fadillah, S. A. N., Mutiarani, A., and Kusumawaty, D. (2022). The effectiveness of mugwort leaf extract and gotu kola leaf extract against acne bacterial activity. *ASEAN Journal of Science and Engineering*, *2*(3), 247-254.

Sari, E., Nugroho, A. P., Retnaningrum, E., and Prijambada, I. D. (2023). Literature review and experiment: Diversity of bacteria in forest, revegetated post-mining land, and active tin mining with a metagenomic approach. *Indonesian Journal of Science and Technology*, *8*(1), 19-48.

Schillinger, U., and L cke, F. K. (1989). Antibacterial activity of Lactobacillus sake isolated from meat. *Applied and Environmental Microbiology, 55*(8), 1901-1906.

Theocharidi, N. A., Balta, I., Houhoula, D., Tsantes, A. G., Lalliotis, G. P., Polydera, A. C., and Halvatsiotis, P. (2022). High prevalence of Klebsiella pneumoniae in Greek meat products: Detection of virulence and antimicrobial resistance genes by molecular techniques. *Foods, 11*(5), 708.

Valgas, C., Souza, S. M. D., Smânia, E. F., and Smânia Jr, A. (2007). Screening methods to determine antibacterial activity of natural products. *Brazilian journal of microbiology*, *38*, 369-380.

Wang, Y., Wu, J., Lv, M., Shao, Z., Hungwe, M., Wang, J., and Geng, W. (2021). Metabolism characteristics of lactic acid bacteria and the expanding applications in food industry. *Frontiers in Bioengineering and Biotechnology*, *9*, 612285.

Wardhana, DK., Haskito, A. E. P., Purnama, M. T. E., Safitri, D. A., and Annisa S. (2021). Detection of microbial contamination in chicken meat from local markets in Surabaya, East Java, Indonesia. *Veterinary World*, *14*(12), 3138-3143.