



Research article

Antifungal activity of lactic acid bacteria from *Pangasius* catfish culture in the Mekong Delta, Vietnam

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Abstract

Fungi are one of the most common pathogens in aquaculture. The study was performed to isolate and select lactic acid bacteria (LAB) with antifungal activity. A total of 36 isolates of LAB were isolated from the intestines of *Pangasianodon hypophthalmus*, water and sludge, fermented vegetables, and “com me” in Vinh Long province of the Mekong Delta, Vietnam. The results revealed that the isolated LAB isolates were inhibitory against five species of fungi: *Aspergillus* sp., *Fusarium* sp., *Achlya* sp., *Saprolegnia* sp., and *Mucor* sp. In particular, isolate LN12 showed the highest activity against *Aspergillus* sp., *Fusarium* sp., *Achlya* sp., *Saprolegnia* sp., and *Mucor* sp. with inhibition diameters of 20 mm, 19.33 mm, 19.67 mm, 19.0 mm, and 20.33 mm, respectively. An investigation on the influence of culture conditions showed that all four bacterial isolates, LN5, LN12, LN23, and LN31, had the highest antifungal activity at a concentration of 10^8 CFU/ml after incubation for 48-60 hours, incubation temperature of 30–35°C, pH of 5.0–6.0 in the medium without the addition of NaCl. Four bacterial isolates, LN5, LN12, LN23, and LN31, were identified as *Lactobacillus*, and LN33 belonged to *Pediococcus* based on morphological and biochemical characteristics, and 16S-rRNA gene sequencing. The results show the potential application of LAB isolates in the prevention of fungal diseases in catfish.

Keywords: Antifungal activity, Fungi, Lactic acid bacteria, Mekong Delta, *Pangasius*

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INTRODUCTION

Pangasius (*Pangasianodon hypophthalmus*) is widely cultivated in the Mekong Delta due to its high economic value. Production of the tra catfish was 1.7 million tons and represented 26% of Vietnam's total value of seafood exports (VASEP, 2022). In addition to bacterial pathogens, previous research has determined that fungi are common pathogenic agents in cultured *Pangasius* (Pham Minh Duc et al., 2010). Research by Dang Thuy Mai Thy et al. (2016) identified five fungal strains commonly infecting *Pangasius* fingerlings: *Aspergillus* sp., *Fusarium* sp., *Achlya* sp., *Saprolegnia* sp., and *Mucor* sp., especially *Fusarium* sp., which seriously causes swollen swim bladder disease in catfish (Duc et al., 2015).

Up to now, the solutions to prevent and treat fungal diseases of aquatic animals in the world and in Vietnam have mainly used drugs and antifungal chemicals (Oono et al., 2008; Duc and Tuan, 2011). However, chemicals and drugs are currently not strictly controlled and managed, so their use can have bad effects on consumers' health, pollute the cultured pond environment, especially because the persistence and release of chemicals to the environment around the aquaculture area have affected the growth of other organisms (de la Casa-Resino et al., 2021; Roy et al., 2021).

Numerous investigations showed that lactic acid bacteria (LAB) were able to inhibit the growth of harmful bacteria (Nurhikmayani et al., 2019; Jawan et al., 2019; Erdoğan et al., 2021; Sharma and Bajwa, 2021). LAB often produce substances such as hydrogen peroxide and organic acids, especially bacteriocins, that can prevent the growth of other pathogenic microorganisms (Karthikeyan and Santosh, 2009). Many studies have demonstrated that the use of LAB increases inhibition and boosts aquatic animal survival rates (Karthikeyan and Santosh, 2009; Meidong et al., 2017; Chizhayeva et al., 2022; Liu et al., 2022). Until now, most of the findings have concentrated on isolating and investigating the inhibitory activity of LAB against pathogenic bacteria on catfish in Vietnam (Thanh and Trai, 2012; Tran et al., 2016). In the meantime, little research has been done on the use of LAB with antifungal activity on *Pangasius* catfish in the Mekong Delta, Vietnam. Therefore, it is crucial to isolate and screen indigenous LAB for fungal inhibitory action.

MATERIALS AND METHODS

Sampling and isolation of LAB

Pangasius catfish, water, and mud samples from *Pangasius* ponds were collected in Long Ho, and Binh Tan districts of Vinh Long province (Figure 1). Each sampling site collected 3–5 fish per pond, weighing between 250 and 870 g. For pond water and sludge samples, collected at different locations on the same pond (5 sites/pond). In addition, some traditional fermented products, such as fermented vegetables and “com me”, a kind of traditional fermented food made from cooked rice in Vietnam, were also obtained from these two sites and used to isolate LAB.

The LAB was isolated from the collected materials using the method described by Vijayabaskar and Somasundaram (2008). In brief, the fish's intestines were homogenized in a 0.9% physiological saline solution, centrifuged at 13,000 rpm per minute for 10 minutes. Then, the supernatant (1.0 ml) was transferred into 9.0 ml of MRS medium (De Man et al., 1960) supplemented with 0.5% CaCO₃, anaerobically incubated at 37°C for 24–48 hours. Finally, the presumptive LAB colonies (Gram-positive, negative-catalase and oxidase reactions) were selected and subcultured on MRS medium many times until the colonies were pure.

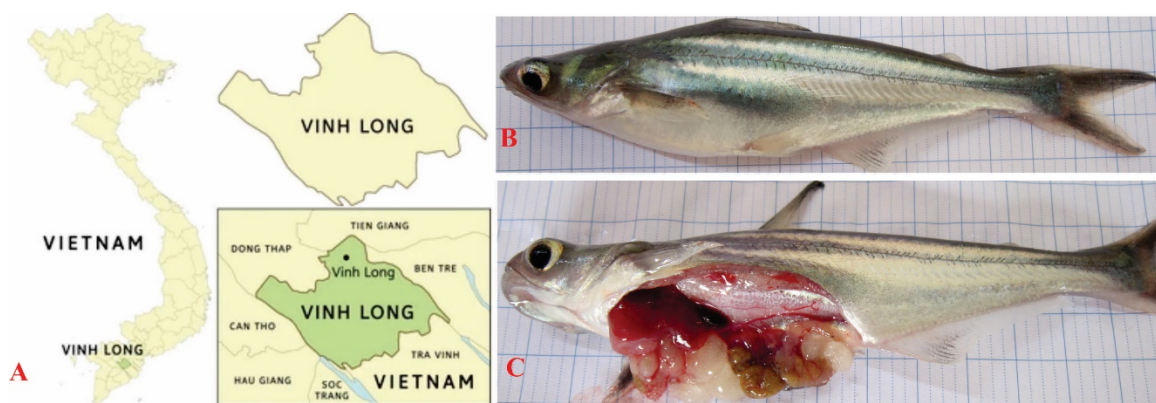


Figure 1 Sampling sites and healthy *Pangasius* sample for LAB isolation
A. Sampling sites (red circle); B. External signs of healthy fish; C. Internal signs of healthy fish

Antifungal activity of LAB isolates

The antifungal activity of LAB in the study was carried out according to Magnusson and Schnurer (2001) with some modifications. Five microfungi species are used as indicators, consisting of *Aspergillus* sp., *Achlya* sp., *Fusarium* sp., *Saprolegnia* sp., and *Mucor* sp. (isolated from diseased catfish). In brief, LAB will be inoculated as two 2-cm-long lines on MRS agar plates. The plates were then covered with 10 ml of PDA, which contained 10^6 fungal spore per mL and was prepared according to the method of Magnusson et al. (2003). Based on the clear zones of inhibition around the bacterial streaks, the antifungal activity of LAB was assessed and graded as follows: –, no inhibition; +, no fungal growth on 0.1 to 3% of the plate area per bacterial streak; ++, no fungal growth on 3 to 8% of the plate area per bacterial streak; or +++, no fungal growth on >8% of the plate area per bacterial streak (Magnusson and Schnurer, 2001).

The antifungal activity of LAB was also examined using the agar well diffusion technique (Magnusson and Schnurer, 2001). Briefly, on the PDA agar plates, fungal spores (10^6 spores/ml) were spread. The base of a sterile pipette tip was then used to make 6 mm-diameter holes in the agar plate. Each well received 80 μ l of each LAB isolate's supernatant. The clearing zone around the wells was then measured after the plate had been incubated aerobically at 30°C for 24 hours. Following are the antifungal activity measurements and grades: -, no inhibition; +, weak inhibition; ++, moderate inhibition; +++, high inhibition.

LAB bacterial identification

LAB isolates were examined for morphological, and biochemical characteristics such as Gram stain, motility, oxidase, and catalase reactions according to Buller (2014). Then, bacteria were identified by using the Lac1 and Lac2 primers with the following sequences: Lac1: 5'-AGCATAGGGAATCTTCCA-3' and Lac2: 5'-ATTCCACCGCTACCATG-3' (Walter, 2001). PCR reaction components include: 1X PCR buffer; 1.5 mM $MgCl_2$; 200 μ M dNTPs; 1.5U *Taq* DNA polymerase; 20 pmol of Lac1 and Lac2 primers; and 40-60 ng of DNA samples. PCR thermal cycling was performed, including initial denaturation at 95°C for 5 minutes, then 30 cycles of denaturation at 95°C for 1 minute, annealing at 55°C for 1 minute, extension at 72°C for 2 minutes, and a final elongation at 72°C for 10 minutes. *L. plantarum* strain RP11-1 (provided by the Faculty of Aquatic Pathology, College of Aquaculture and Fisheries, Can Tho University) was used as reference in this PCR.

The PCR product with a 340 bp size was delivered to Macrogen, Korea (www.macrogen.com), for sequencing. The Clustal W program was used to compare the bacterial sequences to one another (Thompson et al., 1997). Using

bootstrap values of 1.000 replications (Tamura et al., 2013), the phylogenetic tree was created using the MEGA6 (Molecular Evolutionary Genetics Analysis) program, which is based on the neighbor-joining algorithm (Saitou and Nei, 1987).

Effect of culture conditions on antifungal activity

To determine the impact of culture conditions, such as bacterial densities, pH, incubation temperature, incubation period, and NaCl, on their antifungal efficacy for *Aspergillus* sp., *Fusarium* sp., *Achlya* sp., *Saprolegnia* sp., and *Mucor* sp., four isolates, LN5, LN12, LN23, and LN31 were chosen for this investigation.

The influence of bacterial densities

In the experiment, bacterial densities included: 10^4 , 10^5 , 10^6 , 10^7 , and 10^8 CFU ml⁻¹. Bacterial density was determined by measuring optical density (OD) at 600 nm (OD = 1.0, equivalent to a bacterial density of 10^8 CFU ml⁻¹). The antifungal activity of LAB was tested by the agar well diffusion assay (Magnusson and Schnurer, 2001) as described above. The studies were carried out in triplicate using a completely randomized design (CRD), with no bacteria present in the control treatment.

Effect of incubation period

For the study, incubation periods of 24, 36, 48, 60, 72, and 84 hours at 30°C were chosen. The antifungal activity of LAB was achieved in the manner previously mentioned. The studies were carried out in triplicate using a completely randomized design (CRD), with no bacteria present in the control treatment.

Effect of incubation temperature

To explore the antifungal activity of LAB, the incubation temperature values for the survey include: 15°C, 20°C, 25°C, 30°C, and 35°C. The antifungal activity of LAB was achieved in the manner previously mentioned. The studies were carried out in triplicate using a completely randomized design (CRD), with no bacteria present in the control treatment.

Effect of pH

In this experiment, the pH values of 4.0, 5.0, 6.0, 7.0, 8.0, and 9.0 will be investigated. To bring the bacterial culture's pH values up to the appropriate levels, use NaOH or HCl 0.01N. The antifungal activity of LAB was achieved in the manner previously mentioned. The studies were carried out in triplicate using a completely randomized design (CRD), with no bacteria present in the control treatment.

Effect of NaCl

Bacteria were cultured in 2 mL of MRS medium with a bacterial density of 10^8 CFU/ml supplemented with 0% NaCl, 0.5%, 1.0%, 1.5%, 2.0%, and 2.5% to investigate the antifungal activity of LAB. The antifungal activity of LAB was accomplished as described above. The studies were carried out in triplicate using a completely randomized design (CRD), with no bacteria present in the control treatment.

Statistical analysis

The mean ± standard deviation of the antifungal activity of LAB isolates are shown. Using the Minitab 20.0 version, analysis of one-way variance (ANOVA) with Tukey's test was used to identify significant differences between mean values ($P < 0.05$).

RESULTS

Bacterial isolation

The research recovered a total of 36 bacterial isolates on MRS medium from 40 samples collected in two districts, Long Ho and Binh Tan, of Vinh Long province (Table 1). Among the isolates, bacterial isolates derived from fermented vegetables accounted for the highest proportion (12/36 isolates, 33.33%), followed by strains originated from *Pangasius* (10/36 isolates, 27.78%), “com me” (7/36 isolates, 19.44%), and *Pangasius* farming ponds (4/36 isolates, 11.11%), and the lowest was bacteria isolated from bottom mud samples of catfish ponds (3/36 isolates, 8.33%).

Table 1 Isolation of LAB in Long Ho and Binh Tan districts

Isolation source	Sampling location	Number of samples	Number of bacterial isolates	Percentage (%)
Pangasius' intestine	Long Ho	10	6	16.67
	Binh Tan	10	4	11.11
Pond water	Long Ho	3	2	5.56
	Binh Tan	2	2	5.56
Sludge	Long Ho	3	1	2.78
	Binh Tan	2	2	5.56
Fermented vegetables	Long Ho	2	5	13.89
	Binh Tan	3	7	19.44
“Com me”	Long Ho	3	4	11.11
	Binh Tan	2	3	8.33
Total		40	36	100

Antifungal activity of bacterial isolates

The results showed that 14/36 isolates (38.89%) had antifungal activity against *Fusarium* sp. For *Aspergillus* sp., the findings revealed that 17/36 bacterial isolates (47.22%) had inhibitory activity, of which isolate LN12 had the strongest inhibitory activity (Table 2). Meanwhile, the rates of inhibition for *Achlya* sp., *Saprolegnia* sp., and *Mucor* sp. were, respectively, 22/36 isolates (61.11%), 19/36 (52.78%), and 24/36 isolates (66.67%).

Morphological and biochemical characteristics of bacterial colonies

The results showed that most of the colonies of bacterial isolates isolated on MRS medium had a rod (34/36 isolates, 94.44%) or cocci shape (2/36 isolates, 5.56%), milky white, clear white, or yellow color. When growing on MRS agar supplemented with CaCO₃, a clearance zone appeared around the LAB colonies. The biochemical characteristics of all isolates revealed that they were Gram-positive, non-spore-forming, nonmotile, catalase- and oxidase-negative (Table 3).

Table 2 Antifungal activity of bacterial isolates

Isolates	Fungal isolates				
	<i>Aspergillus</i> sp.	<i>Fusarium</i> sp.	<i>Achlya</i> sp.	<i>Saprolegnia</i> sp.	<i>Mucor</i> sp.
LN1	+	+++	++	++	++
LN2	-	-	-	-	+
LN3	-	+	++	++	++
LN4	+++	-	+	-	-
LN5	++	++	+++	+++	+++
LN6	+	++	+	-	-
LN7	-	-	-	-	++
LN8	-	-	-	-	++
LN9	-	-	-	-	++
LN10	++	+	++	++	+
LN11	-	-	+	+	-
LN12	+++	+++	+++	+++	+++
LN14	+	+	++	++	++
LN15	++	-	++	++	++
LN17	-	-	+	-	+
LN18	+++	+	+++	-	++
LN21	++	++	+++	+++	+++
LN22	-	-	++	++	++
LN23	++	+++	+++	+++	+++
LN24	-	-	-	++	++
LN25	-	-	++	++	++
LN26	-	-	+	-	-
LN27	-	-	++	++	++
LN29	++	++	+++	++	+++
LN31	++	+++	+++	+++	+++
LN33	++	++	++	++	++
LN34	+++	-	-	+	-
LN35	+++	-	-	-	++
LN36	++	+++	+++	++	+++

-, no inhibition; +, weak inhibition; ++, moderate inhibition; +++, high inhibition.

Table 3 Morphological and biochemical characteristics of five bacterial isolates LN5, LN12, LN23, LN31, and LN33

Characteristics	Isolate LN5	Isolate LN12	Isolate LN23	Isolate LN31	Isolate LN33	Isolate RP11-1*
Colony color	Yellow	Milky white	Clear white	Milky white	Milky white	Milky white
Gram stain	+	+	+	+	+	+
Spore stain	no	no	no	no	no	no
Cell shape	Long rod	Short rod	Short rod	Short rod	Cocci	Long rod
Motility	no	no	no	no	no	no
Oxidase	-	-	-	-	-	-
Catalase	-	-	-	-	-	-
O/F reaction	+/+	+/+	+/+	+/+	+/+	+/+

* *L. plantarum* strain RP11-1 reference; -: negative; +: positive

Molecular identification of LAB isolates

LAB bacterial isolates with inhibitory activity against five fungal isolates were selected for PCR reactions. The results of PCR product electrophoresis showed that all the bacterial isolates in the study showed a DNA band at 340 bp in size (Figure 2).

Five bacterial isolates (LN5, LN12, LN23, LN31, and LN33) with strong antifungal activity or simultaneous inhibition of multiple fungal isolates were selected for sequencing. Sequencing result demonstrated that the isolates were higher than 99%, closely related to *Lactobacillus*, and *Pediococcus*. The phylogenetic tree showed that the five isolates belonged to two genera (*Lactobacillus*, and *Pediococcus*), and the bacterial isolates were distributed into four separate clusters (Figure 3).



Figure 2 Agarose gel electrophoresis results of PCR products by SafeView DNA stain for detection of LAB isolates

M: 100 bp DNA marker; Lane 1: negative control; Lane 2: positive control (*L. plantarum* strain RP11-1); Lanes 3–16: bacterial isolates LN1, LN3, LN5, LN10, LN12, LN14, LN15, LN18, LN21, LN23, LN29, LN31, LN33, and LN36, respectively.

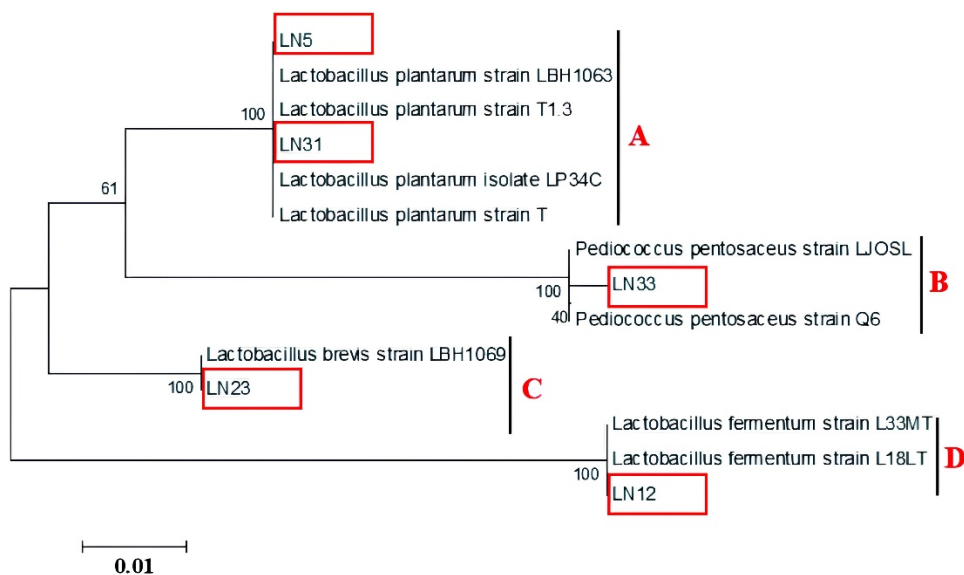


Figure 3 Phylogenetic tree of four LAB isolates based on partial 16S rRNA gene sequences (bootstrap values are given at branching points)
 Cluster A: consist of isolate LN5 and LN31; Cluster B: isolate LN33; Cluster C: isolate LN23; Cluster D: isolate LN12.

Effect of culture conditions on antifungal activity

Impact of bacterial density on the antifungal activity

The findings revealed that the four bacterial isolates' antifungal activity increased with bacterial density. All four isolates displayed the highest antifungal activity in the treatment, with a bacterial density of 10^8 CFU ml⁻¹ (Figure 4). The analysis results revealed a statistically significant difference ($P < 0.05$) in bacterial densities on the antifungal activity at the 5% significance level (Figure 4).

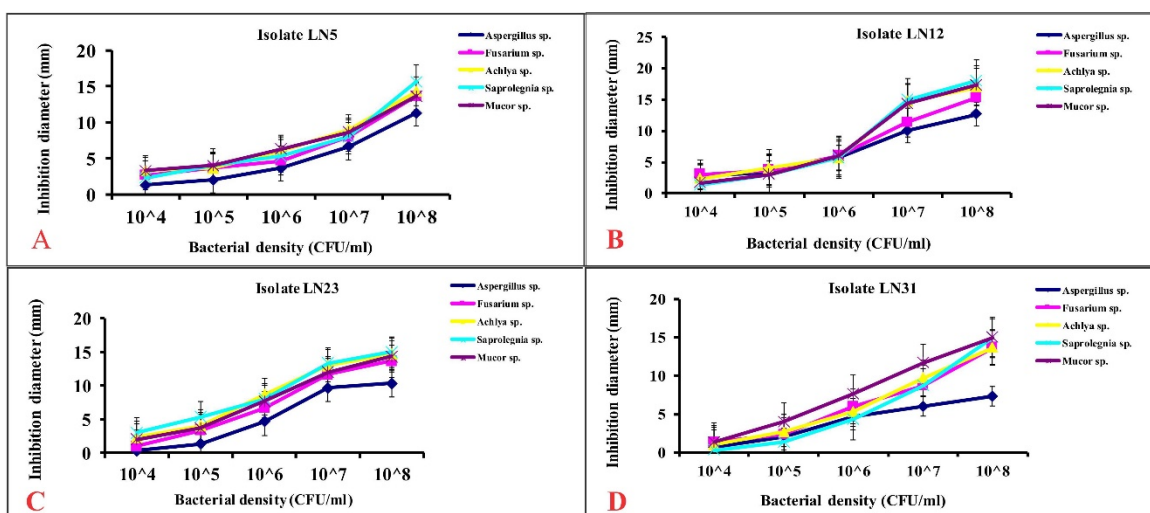


Figure 4 The influence of bacterial density on the antifungal activity of bacterial isolates A. Isolate LN5; B. Isolate LN12; C. Isolate LN23; D. Isolate LN31

Influence of incubation time on the antifungal activity

The findings demonstrated that the isolate LN12 had the highest inhibitory activity at 48 hours after inoculation, while the isolate LN5 and LN23 had the highest inhibition at 60 hours. Meanwhile, strain LN31 had the highest inhibition at different incubation times (Figure 5). In addition, the results also showed that all bacterial isolates showed weak antifungal activity at 24 and 36 hours after incubation. On the other hand, the results also recorded that all bacterial isolates had a decrease in antifungal activity after 60 hours of incubation, with the lowest antifungal activity recorded at 84 hours. At the 5% significance level, the analysis results revealed a statistically significant difference ($P < 0.05$) in incubation time on the antifungal action of four bacterial isolates (Figure 5).

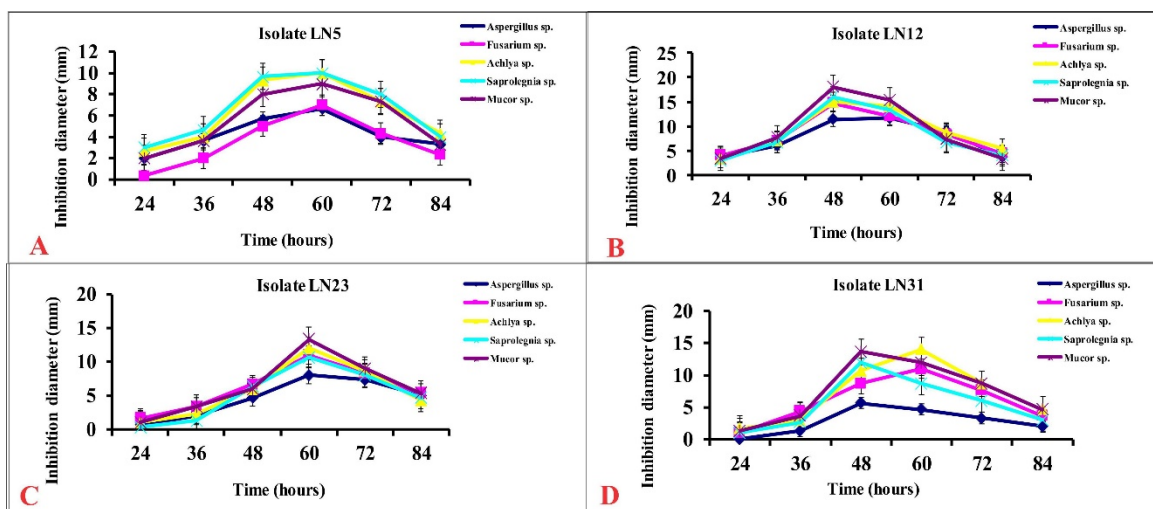


Figure 5 The impact of incubation time on the antifungal activity of bacterial isolates A. Isolate LN5; B. Isolate LN12; C. Isolate LN23; D. Isolate LN31

Impact of incubation temperature on the antifungal activity

The research revealed that at low temperatures (15–20°C), the antifungal activity against five fungal strains was weak (four isolates had weak or no inhibition activity). In general, the antifungal activity of the four bacterial isolates increased sharply at temperatures of 25–35°C. The antifungal activity of the four bacterial isolates was highest at 35°C (Figure 6). However, isolate LN31 was inhibitory against *Aspergillus* sp. at its highest level at 30°C (Figure 6), while *Achlya* sp., *Fusarium* sp., *Saprolegnia* sp., and *Mucor* sp. had the highest inhibitory activity at 35°C. The analysis results showed a statistically significant difference ($P < 0.05$) in the impact of incubation temperature on the antifungal activity of four bacterial strains at the 5% significance level (Figure 6).

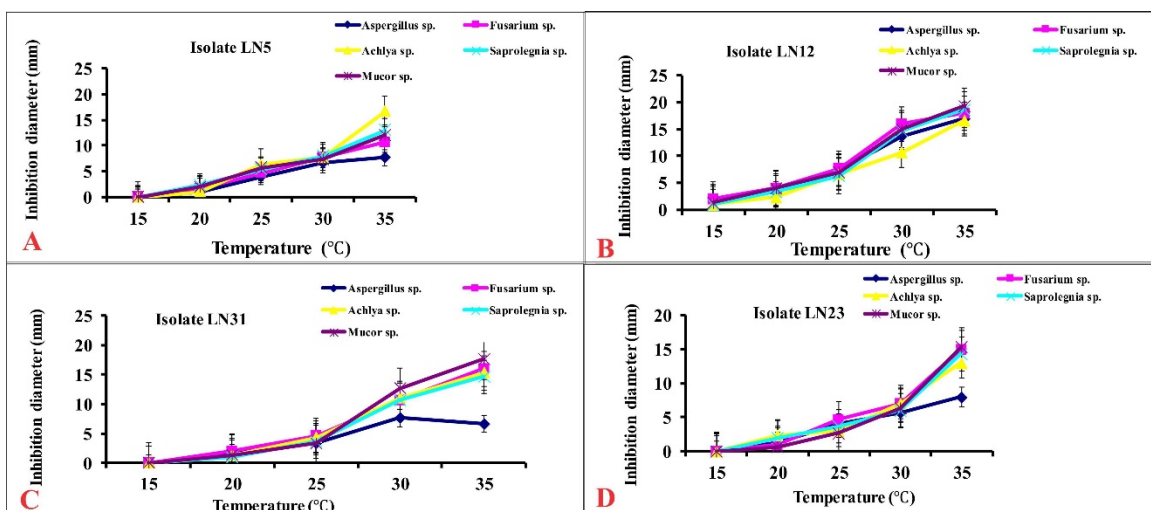


Figure 6 The influence of incubation temperature on the antifungal activity of bacterial isolates A. Isolate LN5; B. Isolate LN12; C. Isolate LN23; D. Isolate LN31

Influence of pH on the antifungal activity

The study demonstrated that the suitable pH for the highest antifungal bacteria was at pH = 5.0 (isolate LN5) and pH = 6.0 (isolate LN12). In general, the higher the pH value, the lower the antifungal activity of the four bacterial isolates, with the lowest antifungal activity at pH = 9.0 (most bacterial isolates have weak or no antifungal activity at this pH value). While at pH = 4.0, the bacterial isolates exhibited moderate prohibitory activity. The analysis results revealed a statistically significant difference ($P < 0.05$) in pH values on the antifungal activity of four bacterial isolates against five fungal isolates at the 5% significance level (Figure 7).

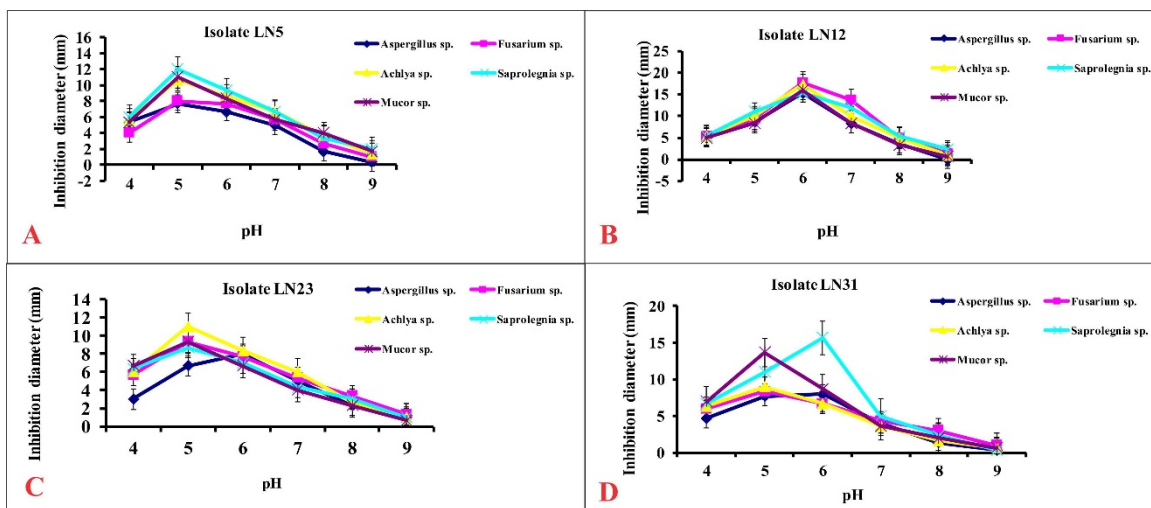


Figure 7 The impact of pH values on the antifungal activity of bacterial isolates A. Isolate LN5; B. Isolate LN12; C. Isolate LN23; D. Isolate LN31

Effect of NaCl on the antifungal activity

The study showed that the majority of bacterial strains showed the highest prohibition against five fungal strains when the medium was not supplemented with NaCl (Figure 8). In general, the experiment revealed that as NaCl concentration

increased, the antifungal activity of the four bacterial isolates decreased. In the treatment with a salt concentration of 2.5%, the antifungal activity was lowest (Figure 8). The results of the analysis showed that there was a statistically significant difference ($P < 0.05$) on the effect of salt concentration on the antifungal activity of four bacterial isolates at the 5% significance level (Figure 8).

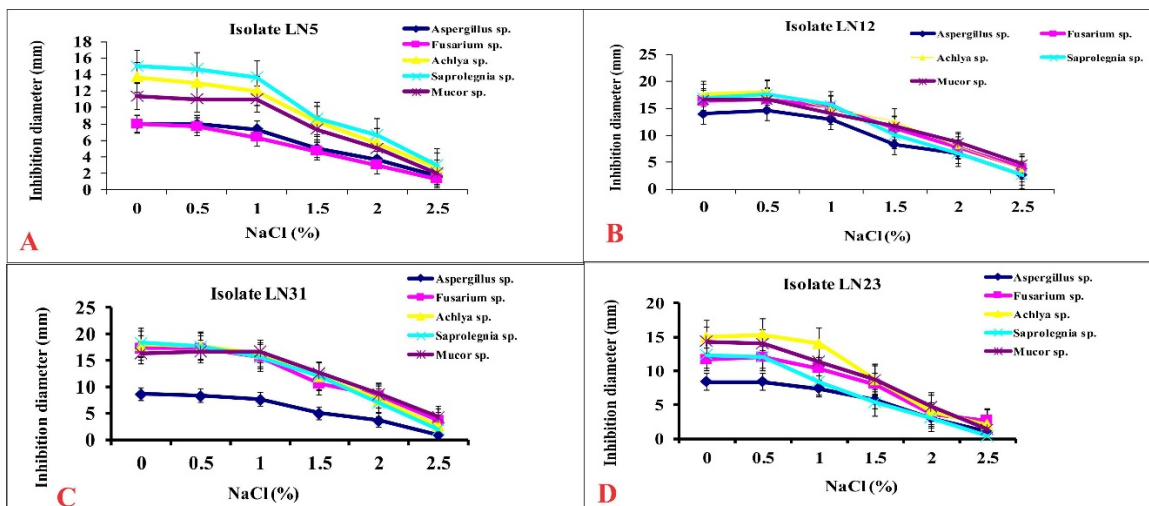


Figure 8 The effect of NaCl on the antifungal activity of bacterial isolates A. Isolate LN5; B. Isolate LN12; C. Isolate LN23; D. Isolate LN31

DISCUSSION

The LAB isolates in the findings were recorded with common morphological characteristics such as round-shaped colonies, milky white, clear white, or light yellow. Besides, all LAB isolates in this study were Gram-positive, non-spore-forming, non-motile, rod-shaped or cocci, and oxidase and catalase-negative bacteria (Table 3). In general, the morphological and biochemical characteristics of LAB in the study were similar to those of LAB reported by many previous authors (Rattanachaikunsopon and Phumkhachorn, 2010; Bourouni et al., 2015; Bintsis, 2018; Sulmiyati et al., 2018). LAB is commonly distributed in traditional fermented products such as fermented vegetables (Tamang et al., 2009), yogurt (Kermanshahi and Peymanfar, 2012; Chen et al., 2017), “com me” (Tran Ngoc Duoc et al., 2013), fermented meats (Parlindungan et al., 2021), and fermented dairy products (Colombo et al., 2018). The findings showed that LAB isolates were also collected from *Pangasius* intestines, water and mud from catfish ponds, fermented vegetable samples, and “com me” in Vinh Long province (Table 1). Research by Yang et al. (2005) isolated 92 strains of LAB from various types of pickles, four of which were identified as *Leuconostoc mesenteroides* subsp. *mesenteroides*, *L. casei* subsp. *casei*, and *L. plantarum*. Yu et al. (2012) isolated 185 strains of LAB from 36 sour cucumber samples, and the identification results indicated that all isolates belonged to *Enterococcus thailandicus* species, *L. alimentarius*, *L. brevis*, *L. paracasei*, *L. plantarum*, *L. pentosus*, *L. sakei*, *L. spcheri*, *Lactococcus lactis*, and *Pediococcus ethanolidurans*.

Many studies have shown that LAB such as *Lactobacillus*, *Streptococcus*, *Leuconostoc*, and *Carnobacterium* belong to the normal microflora in healthy fish guts without harming the host (Ringo and Gatesoupe, 1998; Gatesoupe, 2008; Iorizzo et al., 2021). Jini et al. (2011) isolated LAB species of two genera, *Pediococcus* and *Enterococcus*, from the viscera of many freshwater fish species, such as *Labeo rohita*, *Cirrhinus mrigala*, *Catla catla*, *Cyprinus carpio*, and

Oreochromis mossambicus. The present research also isolated 10 of 36 bacterial isolates from 20 samples of *Pangasius catfish* in Vinh Long province of the Mekong Delta. Similarly, numerous LAB strains, for instance *L. suntuoryeus*, *Enterococcus faecium*, *L. fermentum*, *L. plantarum*, and *L. reuteri*, have also been isolated from many other species of farmed fish, including tilapia (Lara-Flores et al., 2013; Zapata, 2013), *Clarias* sp. (Hamid et al., 2012; Turnip et al., 2018), rainbow trout (Pérez-Sánchez et al., 2011), and the Mediterranean Trout (*Salmo macrostigma*) (Iorizzo et al., 2021).

The most prevalent genus in the LAB group is *Lactobacillus*. In general, this genus has been isolated in the majority of previous studies (Gurovic et al., 2014; Gupta et al., 2019; Rodpai et al., 2021). In this investigation, most collected isolates (34/36 isolates, 94.44 %) were rod-shaped revealed that these bacterial isolates belong to *Lactobacillus* (Wafula et al., 2023). In this study, the result of gene sequencing indicated that isolates LN5, LN12, LN23, and LN31 were highly homologous (>90%) with *L. plantarum*, *L. brevis*, and *L. fermentum* in the NCBI database. Animals with gastrointestinal tracts that contain *Lactobacillus* include cats, pigs, chickens, dogs, and humans (Riaz et al., 2010; Tinrat et al., 2011); in fermented foods such as fermented vegetables, yogurt, cheese, and fermented fish (Rezac et al., 2018; Ngasotter et al., 2020). Additionally, the *L. plantarum* strains were discovered in the intestine of *Penaeus monodon* according to the isolation results of Karthikeyan and Santosh (2009). Furthermore, the study of Kumar et al. (2013) isolated 19 strains of *L. plantarum* from the intestines of freshwater fish such as *Labeo* sp., *Clarias* sp., *Mystus* sp., and *Clupisoma* sp. Meanwhile, under a microscope, 2/36 isolates (5.56%), such as isolate LN33, had cocci shapes. This isolate can be classified as one of the genera *Streptococcus*, *Pediococcus*, *Lactococcus*, *Leuconostoc*, *Tetragenococcus*, *Oenococcus*, and *Vagococcus*. Sequencing of isolate LN33's 16S rRNA gene fragment, however, revealed that it was closely related to *P. pentosaceus*. The phylogenetic tree (Figure 3) also demonstrated that isolate LN33 and other *P. pentosaceus* strains in the NCBI database form a distinct cluster. In light of this, it is possible to say that the isolate LN33 found during the study is *P. pentosaceus*. Similar to *L. plantarum*, *P. pentosaceus* is a homofermentative LAB, which is a non-motile, Gram-positive, and non-spore forming bacterium that gives a negative catalase reaction (Porto et al., 2017). According to the majority of previous studies conducted, *Pediococcus* are widely distributed in dairy products, beverages, and fermented foods (Doan and Ay, 2021; Qi et al., 2021). Moreover, *P. pentosaceus* is capable of producing bacteriocins that prevent some Gram-positive and Gram-negative bacteria (Shin et al., 2008).

To date, most studies have shown that species of the genus *Lactobacillus* are considered beneficial bacteria because of their antibacterial abilities and probiotic properties (Shokryazdan et al., 2014; Arshad et al., 2018; Zhang et al., 2020). Besides, many authors have published LAB with antifungal activity (Laref and Guessas, 2013; Matei and Cornea, 2014; Tropcheva et al., 2014; Sevgi and Tsveteslava, 2015; Meena et al., 2022). The current study showed that the isolated LAB were inhibitory against one to five fungal strains (Table 2). Magnusson et al. (2003) investigated the antifungal activity of 1,200 strains of LAB isolated from different media. The results have identified many bacterial strains in the study that have strong activity against many fungal species, such as *A. nidulans*, *A. fumigatus*, *P. commune*, and *F. sporotrichioides* (including the yeast *Rhodotorula mucilaginosa*). In addition, Kim (2005) collected more than 120 strains of LAB from Kimchi, and the study identified five fungal strains that are simultaneously inhibitory to fungi such as *Rhizopus oryzae*, *A. flavus*, *P. commune*, and *F. moniliforme*. In China, Wang et al. (2012) isolated *L. plantarum* IMAU10014 from koumiss (fermented milk). Research results have for the first time identified the antifungal substances benzoic acid and 2-propenyl ester from this bacterium, and they can inhibit many pathogenic fungi such as *P. citrinum*, *Botrytis cinerea*, *Glomerella*

cingulate, *P. digitatum*, *Phytophthora drechsleri* Tucker, and *F. oxysporum*. In particular, the fungal strains *F. oxysporum*, *P. citrinum*, and *P. drechsleri* Tucker are most susceptible to the bacteria. [Tatsadjieu et al. \(2016\)](#) isolated 53 strains of LAB from corn, such as dried corn, soaked corn, and fermented corn paste in Cameroon, that were inhibitory to 21 strains of fungi, including six strains of LAB (including *L. brevis* (two strains), *L. buchneri* (one strain), *L. cellobiosus* (one strain), and *L. fermentum* (two strains) that were resistant to four fungi: *Penicillium*, *Fusarium*, *Aspergillus*, and *Rhizopus*. The highest antifungal activity of isolated LAB bacterial strains shows their potential for application in the production of probiotics to help prevent diseases, stimulate the immune system, and increase fish growth. However, it is necessary to evaluate probiotic characteristics such as hemolytic activity, low pH and bile salt tolerance, adhesion ability, and antibiotic resistance of LAB bacterial strains before recommending them to fish farmers.

The results showed that the incubation times for four bacterial isolates in the study having the highest antifungal activity was shorter than the previous research results ([Figure 5](#)). Research published by [Magnusson and Schnurer \(2001\)](#) showed that *L. coryniformis* subsp. *coryniformis* strain Si3 had the highest antifungal activity at 40 hours. Meanwhile, the report of [Wulijideligen et al. \(2011\)](#) showed that *Leuconostoc mesenteroides* had the highest antifungal activity at 54 and 60 hours. However, the antifungal activity of LAB decreased with longer incubation times ([Batish et al., 1990](#)). In Malaysia, [Muhialdin and Hassan \(2011\)](#) isolated 137 strains of LAB from fermented foods and fruits. In this investigation, the results showed that 23 of the tested fungal strains indicated inhibitory activity at 72 hours, with an incubation time of 30°C. Similarly, the study by [Kivanc et al. \(2014\)](#) also demonstrated that some strains of LAB isolated from tarhana (a traditional fermented food) exhibited inhibitory activity at 72 hours with an incubation temperature of 30°C.

Regarding the pH effect, the study is similar to reports by [Batish et al. \(1997\)](#), which showed that *Lc. lactis* subsp. *diacetyllactis* can produce antifungal agents at a narrow pH range, ranging from 5.5 to 7.0, although the bacteria achieved the highest inhibitory effect at pH = 6.8. Similarly, the study by [Corsetti et al. \(1998\)](#) also showed that *L. sanfrancisco* CB1 had good antifungal activity at pH = 6.0. However, other research results show that LAB have strong antifungal activity at low pH values. According to research done by [Magnusson and Schnurer \(2001\)](#), *L. coryniformis* subsp. *coryniformis* strain Si3 displayed steady antifungal activity at pH levels between 3.0 and 4.5 but significantly declined between 4.5 and 6.0. Furthermore, the results also showed that at pH > 6.0, the bacteria did not have antifungal activity. Research by [Rouse et al. \(2008\)](#) showed that the antifungal activity of *P. pentosaceus* strains was stronger at low pH, although inhibition still occurred at pH = 8.0.

The influence of incubation temperature on the antifungal activity of four bacterial isolates in the study was similar to the previous research. The findings by [Rouse et al. \(2008\)](#) showed that *P. pentosaceus* produced the highest antifungal activity at temperatures between 25°C and 30°C, while bacteria showed no antifungal activity at 10°C and 42°C. Research by [Sathe et al. \(2007\)](#) showed antifungal activity of *L. plantarum* CUK501 was highest at 30°C when the bacterial culture was at the end of the log period. For the effect of NaCl, the findings are similar to those of [Batish et al. \(1989\)](#) showed that a high concentration of NaCl reduces the antifungal activity of *Lc. lactis* subsp. *diacetyllactis*. However, the research by [Effat et al. \(2001\)](#) indicated that when increasing the NaCl concentration from 0.5% to 3% in the growth medium of *Lb. rhamnosus* would increase the antifungal activity of this bacteria.

CONCLUSIONS

The LAB isolated from the intestine of *Pangasius*, water and sludge in *Pangasius* ponds, fermented vegetable samples, and “com me” were inhibitory against five strains of fungi: *Aspergillus* sp., *Achlya* sp., *Fusarium* sp., *Mucor* sp., and *Saprolegnia* sp. Four isolates of LN5, LN12, LN23, and LN31 had the highest antifungal activity at a density of 10^8 CFU ml⁻¹ after incubation of 48–60 hours, at a temperature of 30–35°C, and a pH of 5.0–6.0 without the supplementation of NaCl. The results show the potential application of LAB isolates in the prevention of fungal diseases in catfish.

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CONFLICT OF INTEREST

The authors have no conflict of interest to declare.

REFERENCES

- Arshad, F.A., Mehmood, R., Hussain, S., Khan, M.A., Khan, M.S., 2018. Lactobacilli as probiotics and their isolation from different sources. *Br. J. Res.* 5(3), 1-11.
- Batish, V.K., Lal, R., Grove, S., 1989. Screening lactic acid starter cultures for antifungal activity. *J. Cult. Dairy. Prod.* 24(2), 21-25.
- Batish, V.K., Lal, R., Grover, S., 1990. Effect of nutritional factors on the production of antifungal substance by *Lactococcus lactis* biovar Diacetylactis. *Aust. J. Dairy. Technol.* 45(2), 74-76.
- Batish, V.K., Roy, U., Lal, R., Grover, S., 1997. Antifungal attributes of lactic acid bacteria – A review. *Crit. Rev. Biotechnol.* 17(3), 2009-2225.
- Bintsis, T., 2018. Lactic acid bacteria as starter cultures: an update in their metabolism and genetics. *AIMS Microbiol.* 4(4), 665–684.
- Bourouni, O.C., Barros-Velázquez, J., Calo-Mata, P., El Bour, M., 2016. Lactic acid bacteria associated with the digestive tract and skin of Sea bream (*Sparus aurata*) cultured in Tunisia. *Afr. J. Microbiol. Res.* 9(31), 1853-1865.
- Buller, N.B., 2014. Bacteria and fungi from fish and other aquatic animals: a practical identification manual, 2nd edition. CABI, Wallingford.
- Chen, C., Zhao, S., Hao, G., Yu, H., Tian, H., Zhao, G., 2017. Role of lactic acid bacteria on the yogurt flavour: a review. *Int. J. Food. Prop.* 20(S1), S316-S330.
- Chizhayeva, A., Amangeldi, A., Oleinikova, Y., Alybaeva, A., Sadanov, A., 2022. Lactic acid bacteria as probiotics in sustainable development of aquaculture. *Aquat. Living Resour.* 35(10), 1-17.
- Colombo, M., Castilho, N.P.A., Todorov, S.D., Nero, L.A., 2018. Beneficial properties of lactic acid bacteria naturally present in dairy production. *BMC Microbiol.* 18(219), 1-12.
- Corsetti, A., Gobetti, M., Rossi, J., Damiani, P., 1998. Antimould activity of sourdough lactic acid bacteria: identification of a mixture of organic acids produced by *Lactobacillus sanfrancisco* CB1. *Appl. Microbiol. Biotechnol.* 50(2), 253-256.

- de la Casa-Resino, I., Empl, M.T., Villa, S, Kolar, B., Fabrega, J., Lillicrap, A.D., Karamanlis, X.N., Carapeto-García, R., 2021. Environmental risk assessment of veterinary medicinal products intended for use in aquaculture in Europe: the need for developing a harmonised approach. *Environ. Sci. Eur.* 33(84), 1-17.
- De Man, J.C., Rogosa, M., Sharpe, M.E., 1960. A medium for the cultivation of Lactobacilli. *J. Appl. Bacteriol.* 23(1), 130-135.
- Doğan, M., Ay, M., 2021. Evaluation of the probiotic potential of *Pediococcus* strains from fermented dairy product kefir. *Czech J. Food Sci.* 39(5), 376-383.
- Duc, P.M., Tuan, T.N., 2011. In vitro activities of chemicals and antifungal agents against *Plectosporium oratosquillae* and *Acremonium* sp. *Can Tho Univ. J. Sci.* 19b(2011), 20-29
- Duc, P.M., Phuong, N.T., Tuan, T.N., 2010. Overview fungal diseases in aquatic animals. *Can Tho Univ. J. Sci.* 16b(2010), 88-97
- Duc, P.M., Thy, D.T.M., Hatai, K., Muraosa, Y., 2015. Infection of striped catfish (*Pangasianodon hypophthalmus*) in Viet Nam caused by the fungus *Fusarium incarnatum-equiseti*. *Bull. Eur. Ass. Fish. Pathol.* 35(6), 208-216.
- Duoc, T.N., Dung, T.N., Dieu, B.T.M., Khang, D.T., 2013. Diversity of lactic acid bacteria isolated from “Com me” collected from different ecological regions in Mekong Delta. *Can Tho Univ. J. Sci.* 25(2013), 58-66.
- Effat, B.A., Ibrahim, G.A., Tawfik, N.F., Sharaf, O.M., 2001. Comparison of antifungal activity of metabolites from *Lactobacillus rhamnosus*, *Pediococcus acidilactici* and *Propionibacterium thoenii*. *Egypt. J. Dairy. Sci.* 29(2), 251-262.
- Erdoğmuş, S.F., Erişmiş, U.C., Uğuz, C., 2021. Isolation and identification of lactic acid bacteria from fermented meat products and evaluation of their antimicrobial effect. *Czech. J. Food. Sci.* 39(4), 289-296.
- Gatesoupe, F.J., 2008. Updating the importance of lactic acid bacteria in fish farming: natural occurrence and probiotic treatments. *J. Mol. Microbiol. Biotechnol.* 14(1-3), 107-14.
- Gupta, S., Fečkaninová, A., Lokesh, J., Koščová, J., Sørensen, M., Fernandes, J., Kiron, V., 2019. *Lactobacillus* dominate in the intestine of Atlantic salmon fed dietary probiotics. *Front. Microbiol.* 9, 3247.
- Gurovic, M.S.V., Gentili, A.R., Olivera, N.L., Rodriguez, M.S., 2014. Lactic acid bacteria isolated from fish gut produce conjugated linoleic acid without the addition of exogenous substrate. *Process. Biochem.* 49, 1071-1077.
- Hamid, T.H.A.T., Khan, A.J., Jalil, M.F., Azhar, N.S., 2012. Isolation and screening of lactic acid bacteria, *Lactococcus lactis* from *Clarias gariepinus* (African catfish) with potential use as probiotic in aquaculture. *Afr. J. Biotechnol.* 11(29), 7494-7499.
- Iorizzo, M., Albanese, G., Testa, B., Ianiro, M., Letizia, F., Succi, M., Tremonte, P., D'Andrea, M., Iaffaldano, N., Coppola, R., 2021. Presence of lactic acid bacteria in the intestinal tract of the mediterranean trout (*Salmo macrostigma*) in its natural environment. *Life.* 11(7), 667.
- Jawan, R., Kasimin, M., Jalal, S., Mohd Faik, A., Abbasiliasi, S., Ariff, A., 2019. Isolation characterisation and in vitro evaluation of bacteriocins-producing lactic acid bacteria from fermented products of Northern Borneo for their beneficial roles in food industry. *J. Phys. Conf. Ser.* 1358, 012020.
- Jini, R., Swapna, H.C., Rai, A.K., Vrinda, R., Halami, P.M., Sachindra, N.M., Bhaskar, N., 2011. Isolation and characterization of potential lactic acid bacteria (LAB) from freshwater fish processing wastes for application in fermentative utilisation of fish processing waste. *Braz. J. Microbiol.* 42(4), 1516-1525.
- Karthikeyan, V., Santosh, S.W., 2009. Isolation and partial characterization of bacteriocin produced from *Lactobacillus plantarum*. *Afr. J. Microbiol. Res.* 3(5), 233-239.

- Kermanshahi, R.K., Peymanfar, S., 2012. Isolation and identification of Lactobacilli from cheese yoghurt and silage by 16S rDNA gene and study of bacteriocin and biosurfactant production. *Jundishapur. J. Microbiol.* 5(4), 528-532.
- Kim, J.D., 2005. Antifungal activity of lactic acid bacteria isolated from Kimchi against *Aspergillus fumigatus*. *Mycobiol.* 33(4), 210-214.
- Kivanc, M., Kivanc, S.A., Pektas, S., 2014. Screening of lactic acid bacteria for antifungal activity against fungi. *J. Food. Process. Technol.* 5(3), 310.
- Kumar, Y., Chisti, B., Singh, A.K., Masih, H., Mishra, S., 2013. Isolation and characterization of Lactobacillus species from fish intestine for probiotic properties. *Int. J. Pharma. Bio. Sci.* 4(1), 11-21
- Lara-Flores, M., Olvera-Novoa, M.A., 2013. The use of lactic acid bacteria isolated from intestinal tract of Nile tilapia (*Oreochromis niloticus*) as growth promoters in fish fed low protein diets. *Lat. Am. J. Aquat. Res.* 41(3), 490-497.
- Liu, Y., Lv, H., Xu, L., Zhang, K., Mei, Y., Chen, J., Wang, M., Guan, Y., Pang, H., Wang, Y., Tan, Z., 2022. The effect of dietary lactic acid bacteria on intestinal microbiota and immune responses of crucian carp (*Carassius auratus*) under water temperature decrease. *Front. Microbiol.* 13, 847167.
- Magnusson, J., Schnurer, J., 2001. Lactobacillus coryniformis subsp. coryniformis strain Si3 produces a broad-spectrum proteinaceous antifungal compound. *Appl. Environ. Microbiol.* 67(1), 1-5.
- Magnusson, J., Strom, K., Roos, S., 2003. Broad and complex antifungal activity among environmental isolates of lactic acid bacteria. *FEMS Microbiol. Lett.* 219(1), 129-135.
- Matei, M., Cornea, C.P., 2014. Antifungal activity of some lactic acid bacteria isolated from materials of vegetal origin. *Sci. Bull. F Biotechnol.* 18, 42-47.
- Meena, K.K., Taneja, N.K., Jain, D., Ojha, A., Kumawat, D., Mishra, V., 2022. In vitro assessment of probiotic and technological properties of lactic acid bacteria isolated from indigenously fermented cereal-based food products. *Ferment.* 8(10), 529.
- Meidong, R., Doolgindachbaporn, S., Sakai, K., Tongpim, S., 2017. Isolation and selection of lactic acid bacteria from Thai indigenous fermented foods for use as probiotics in tilapia fish *Oreochromis niloticus*. *AACL Bioflux.* 10(2), 455-463.
- Muhialdin, B.J., Hassan, Z., 2011. Screening of lactic acid bacteria for antifungal activity against *Aspergillus oryzae* American. *J. Appl. Sci.* 8(5), 447-451.
- Ngasotter, S., Waikhom, D., Mukherjee, S., 2020. Diversity of lactic acid bacteria (LAB) in fermented fish products: a review. *Int. J. Curr. Microbiol. Appl. Sci.* 9(05), 2238-2249.
- Nurhikmayani, R., Daryono, B.S., Retnaningrum, E., 2019. Isolation and molecular identification of antimicrobial-producing lactic acid bacteria from chao South Sulawesi (Indonesia) fermented fish product. *Biodivers.* 20(4), 1063-1068.
- Oono, H., Hatai, K., Aikawa, H., Hara, H., 2008. The use of bronopol to control fungal infection in Ayu eggs. *Aquac. Sci.* 56(1), 9-12.
- Parlindungan, E., Lugli, G.A., Ventura, M., van Sinderen, D., Mahony, J., 2021. Lactic acid bacteria diversity and characterization of probiotic candidates in fermented meats. *Foods.* 10(7), 1519.
- Pérez-Sánchez, T., Balcázar, J.L., García, Y., Halaihel, N., Vendrell, D., de Blas, I., Merrifield D.L., Ruiz-Zarzuela, I., 2011. Identification and characterization of lactic acid bacteria isolated from rainbow trout *Oncorhynchus mykiss* (Walbaum) with inhibitory activity against *Lactococcus garvieae*. *J. Fish. Dis.* 34(7), 499-507.
- Porto, M.C.W., Kuniyoshi, T.M., Azevedo, P.O.S., Vitolo, M., Oliveira, R.P.S., 2017. *Pediococcus* spp.: an important genus of lactic acid bacteria and pediocin producers. *Biotechnol. Adv.* 35(3), 361-374.
- Qi, Y., Huang, L., Zeng, Y., 2021. *Pediococcus pentosaceus*: screening and application as probiotics in food processing. *Front. Microbiol.* 12, 762467.

- Rattanachaikunsopon, P., Phumkhachorn, P., 2010. Lactic acid bacteria: their antimicrobial compounds and their uses in food production. *Ann. Biol. Res.* 1(4), 218-228.
- Rezac, S., Kok, C.R., Heermann, M., Hutkins, R., 2018. Fermented foods as a dietary source of live organisms. *Front. Microbiol.* 9, 1785.
- Riaz, S., Nawaz, S.K., Hasnain, S., 2010. Bacteriocins produced by *Lactobacillus fermentum* and *Lactobacillus acidophilus* can inhibit cephalosporin resistant *E. coli*. *Braz. J. Microbiol.* 41(3), 643-648.
- Ringo, E., Gatesoupe, F.G., 1998. Lactic acid bacteria in fish: a review. *Aquac.* 160(3-4), 177-203.
- Rodpai, R., Sanpool, O., Thanchomnang, T., Wangwiwatsin, A., Sadaow, L., Phupiewkham, W., Boonroumkaew, P., Intapan, P.M., Maleewong, W., 2021. Investigating the microbiota of fermented fish products (Pla-ra) from different communities of northeastern Thailand. *PLoS ONE.* 16(1), e0245227.
- Rouse, S., Harnett, D., Vaughan, A., van Sinderen, D., 2008. Lactic acid bacteria with potential to eliminate fungal spoilage in foods. *J. Appl. Microbiol.* 104(3), 915-923.
- Roy, A., Ghosh, S.K., Hauzoukim Swain, S., Bhattacharya, K., Mukherjee, D., Bardhan, A., 2021. Aqua drugs and chemicals used in freshwater aquaculture: a review. *Pharm. Innov. J. SP.* 10(8), 317-324.
- Saitou, N., Nei, M., 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* 4(4), 406-425.
- Sathe, S.J., Nawani, N.N., Dhakephalkar, P.K., Kapadnis, B.P., 2007. Antifungal lactic acid bacteria with potential to prolong shelf-life of fresh vegetables. *J. Appl. Microbiol.* 103(6), 2622-2628.
- Sevgi, E., Tsveteslava, I., 2015. Antifungal Activity of lactic acid bacteria, isolated from Bulgarian Wheat and Rye Flour. *J. Life. Sci.* 9(2015), 1-6.
- Sharma, H., Bajwa, J., 2021. Isolation and characterization of lactic acid bacteria isolated from fermented food of North-West Himalayas. *J. Pure. Appl. Microbiol.* 15(3), 1567-1572.
- Shin, M.S., Han, S.K., Ryu, J.S., Kim, K.S., Lee, W.K., 2008. Isolation and partial characterization of a bacteriocin produced by *Pediococcus pentosaceus* K23-2 isolated from Kimchi. *J. Appl. Microbiol.* 105(2), 331-339.
- Shokryazdan, P., Sieo, C.C., Kalavathy, R., Liang, J.B., Alitheen, N.B., Jahromi, M.F., Ho, Y.W., 2014. Probiotic potential of *Lactobacillus* strains with antimicrobial activity against some human pathogenic strains. *Biomed. Res. Int.* 2014, 927268.
- Sulmiyati Said, N.S., Fahrodi, D.U., Malaka, R., Maruddin, F., 2018. The characteristics of lactic acid bacteria isolated from Indonesian commercial kefir grain. *Malays J. Microbiol.* 14(7), 632-639.
- Tamang, J.P., Tamang, B., Schillinger, U., Guigas, C., Holzapfel, W.H., 2009. Functional properties of lactic acid bacteria isolated from ethnic fermented vegetables of the Himalaya. *Int. J. Food. Microbiol.* 135(1), 28-33.
- Tamura, K., Stecher, G., Peterson, D., Filipowski, A., Kumar, S., 2013. MEGA6: Molecular evolutionary genetics analysis version 6.0. *Mol. Biol. Evol.* 30(12), 2725-2729.
- Tatsadjieu, L.N., Tchikoua, R., Funtong, C.M.M., 2016. Antifungal Activity of lactic acid bacteria against molds isolated from corn and fermented corn paste. *Am. J. Microbiol. Res.* 4(4), 90-100.
- Thanh, N.V., Trai, N.N., 2012. Isolation of *Lactobacillus* sp. inhibiting bacteria causing "red-sore disease" and "white spot in the internal organs" on *Pangasianodon hypophthalmus*. *Can Tho Univ. J. Sci.* 23a(2012), 224-234.
- Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., Higgins, D.G., 1997. The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic. Acids. Res.* 25(24), 4876-4882.

- Thy, D.T.M., Duc, P.M., Hoa, T.T.T., 2016. Identification of filamentous fungi effecting striped catfish fingerling (*Pangasianodon hypophthalmus*). *Can Tho Univ. J. Sci.* 44b(2016), 48-57.
- Tinrat, S., Saraya, S., Traidej, C.M., 2011. Isolation and characterization of *Lactobacillus salivarius* MTC 1026 as a potential probiotic. *J. Gen. Appl. Microbiol.* 57(6), 365-378.
- Tran, N.T.N., Nghia, N.T., Oanh, D.T.H., 2016. Determination of antibacterial character of Lactic acid bacteria to *Streptococcus agalactiae* isolated from haemorrhagic diseased tilapia (*Oreochromis niloticus*). *Can Tho Univ. J. Sci.* 42(2016), 48-55.
- Tropcheva, R., Nikolova, D., Evstatieva, Y., Danova, S., 2014. Antifungal activity and identification of *Lactobacilli*, isolated from traditional dairy product "katak. *Anaerobe.* 28(2014), 78-84.
- Turnip, E.R., Widanarni, Meryandini, A., 2018. Selection of lactic acid bacteria as a probiotic and evaluated its performance on gnotobiotic catfish *Clarias* sp. *J. Akuakultur Indones.* 17(1), 68-80.
- Vietnam Association of Seafood Exporters and Producers (VASEP), 2022. Overview of pangasius industry. Available online: <https://vasep.com.vn/san-pham-xuat-khau/ca-tra/tong-quan-nganh-ca-tra>
- Vijayabaskar, P., Somasundaram, S.T., 2008. Isolation of bacteriocin producing Lactic Acid Bacteria from fish gut and probiotic activity against common fresh water fish pathogen *Aeromonas hydrophila*. *Biotechnol.* 7(1), 124-128.
- Wafula, E.N., Kuja, J.O., Wekesa, T.B., Wanjala, P.M., 2023. Isolation and identification of autochthonous lactic acid bacteria from commonly consumed African indigenous leafy vegetables in Kenya. *Bacteria.* 2(1), 1-20.
- Walter, J., Hertel, C., Tannock, G.W., Lis, C.M., Munro, K., Hammes, W.P., 2001. Detection of *Lactobacillus Pediococcus*, *Leuconostoc* *Weisella* species in human feces by using group-specific PCR primers and denaturing gradient gel electrophoresis. *Appl. Environ. Microbiol.* 67(6), 2578-2585.
- Wang, H., Yan, Y., Wang, J., 2012. Production and characterization of antifungal compounds produced by *Lactobacillus plantarum* IMAU10014. *PLoS ONE.* 7(1), e29452.
- Wulijideligen, S., Miyamoto, T., 2011. Screening and identification of lactic acid bacteria from Airag for antifungal activity. *J. Anim. Vet. Adv.* 10(21), 2751-2757.
- Yang, X.H., Ji, B.P., Li, B., He, S.L., 2005. Isolation, identification and fermentation characteristics study of lactic acid bacteria from pickles. *Food. Sci.* 6(4), 130-134.
- Yu, J., Gao, W., Qing, M., Sun, Z., Wang, W., Liu, W., Pan, L., Sun, T., Wang, H., Bai, N., Zhang, H., 2012. Identification and characterization of lactic acid bacteria isolated from traditional pickles in Sichuan China. *J. Gen. Appl. Microbiol.* 58(3), 163-172.
- Zapata, A.A., Lara-Flores, M., 2013. Antimicrobial activities of lactic acid bacteria strains isolated from Nile Tilapia intestine (*Oreochromis niloticus*). *J. Biol. Life Sci.* 4(1), 164-171.
- Zhang, X., Esmail, G.A., Alzeer, A.F., 2020. Probiotic characteristics of *Lactobacillus* strains isolated from cheese and their antibacterial properties against gastrointestinal tract pathogens. *Saudi J. Biol. Sci.* 27(12), 3505-3513.

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