SELECTION OF LACTIC ACID BACTERIA ISOLATED FROM COCOA AND CASSAVA FERMENTATION AS POTENTIAL PROBIOTIC FOR PATHOGENIC MICROORGANISMS CONTROL IN POULTRY FARMING

ABSTRACT

Aims: The overuse of antibiotics in animal farming sector is leading to an increase drug resistant bacteria rate. This situation makes it difficult to treat pathologies in both humans and animals. The aim of this study was to assess some probiotic profiles of lactic acid bacteria strains isolated from cocoa fermentation and traditional cassava ferment for possible use as potentially probiotic strains for the monitoring of pathogenic microorganisms in poultry farming.

Methodology: Thus, a total of 267 lactic acid bacteria strains were tested for analysis of the antibacterial activity against the growth capacity of *Salmonella* and *E*. *coli* isolates. Probiotic properties of Lactic acid bacteria were consisted of acidification capacity, resistance to acid shock and to salt bile containing in culture medium and capacity to produce proteolytic and lipolytic enzymes.

Results: Among them, 25 strains have induced the high bacterial growth inhibition against these pathogenic bacteria with inhibition zone diameters ranged from 9 to 27 mm. Among these strains, 20 isolates showed high resistance to acid shock at pH ≥ 4 and six strains were able to grow at pH 3.5 with survival rate range from 30 % to 89 %. Moreover, six of these strains, including four isolates of *Lactobacillus plantarum* (T1GB8, T11AB17, LAB26, LAB 127), one strain of *Leuconostoc mesenteroides* (T0AB9) and one isolate of *Enterococcus facium* (LAB18), have shown capacity to growth with 1 % of bile salts in the medium. Even better, these strains exhibited capacity to produce proteolytic and lipolytic enzymes with halo around the well diameters reached 29 mm for some strains.

Conclusion: This study shows the possibility of use probiotics lactic acids bacteria as antibiotics alternative in poultry sector to reduce some avian pathologies affecting the poultry sector in Côte d'Ivoire.

Keywords: Lactic acid bacteria, antibiotics, drugs resistance, avian pathologies

1. INTRODUCTION

In Côte d'Ivoire, the poultry industry is today an important economic activity which contributes to 4.5% of agricultural GDP and 2 % of total GDP [1]. Indeed, the implementation of the Strategic Plan for the Revival of Poultry Farming (PSRA), since 2012, has allowed achieving this performance. In addition, the government intends to increase this production to fully cover the needs of Ivorians poultry meat and achieve self-sufficiency in 2029.

However, poultry production sector is constantly affecting by several illnesses which lead to a significant decrease of chicken meat and eggs production. Indeed, the problem seems to have become more alarming in recent decades; the prevalence of these diseases is increasing, particularly on modern farms. To overcome these problems, farmers are turning to the overuse of antibiotics [2].

Generally, the treatment of these infections relies entirely on an antibiotic therapy [3]. However, in developing countries as Côte d'Ivoire, there is no surveillance system for antibiotic use during breeding.

Consequently, antibiotics are widely using to prevent, control, and treat bacterial infections as well as growth promoters during poultry production [4]. This overuse of several molecules in poultry production systems promotes the development of resistant and even multidrug-resistant bacteria [5]. In this conditions, antibiotic treatment of poultry's microbial diseases in farms becomes inefficacy, resulting in enormous economic losses.

Therefore, several alternatives to antibiotics have been proposed among which the use of probiotics. Probiotics are live microorganisms which, in sufficient quantities, exert a positive effect on health. They play an important role in improving digestion and intestinal transit, maintaining the balance of the intestinal flora and the acid-base balance in the colon.

Lactic acid bacteria currently form a group of organisms were used for the enrichment of certain yogurts and milks [6]. This use is due to the nutritional and therapeutic effects of these bacteria because they enrich the medium with vitamins (B and K), amino acids, organic compounds (lactic and acetic acids), enzymes (lactase) and bacteriocins responsible for the inhibition of pathogenic bacteria [7]. The bacteria most frequently use as probiotics are *Lactobacillus* and *Bifidobacterium* [8]. Lactobacilli have been incorporated in fermented milks [9, 10], cheeses [11,12] and ice creams [13]. In addition, any isolates probiotic-producing bacterial strain should resist a wide variety of conditions such as exposure to digestive enzymes in the oral and gastric cavities, acid pH in the stomach, reduced O_2 content in the intestine, and a temperature that is not always optimal.

Moreover, they are able to inhibit the growth of pathogenic organisms through different mechanisms such as adherence to epithelial cells, modulation of the immune system, and secretion of antimicrobial compounds [14].

The aim of this study was to assess the probiotic profile of lactic acid bacteria strains isolated from cocoa fermentation and traditional cassava ferment for possible use as potentially probiotic strains for the control of pathogenic microorganisms in poultry farming.

2. MATERIAL AND METHODS

2.1 Lactic acid bacteria culture

A total of 267 Lactic acid bacteria from "Magnan" cassava ferment and cocoa beans during fermenting process were used in this study. All strains were previously stored at -80°C in MRS buffer medium contained 20 % (v/v) of glycerol. Thus, each isolate was cultured in 5 ml of sterile de Man, Rogosa, Sharpe (MRS) broth (BioRad, France) and incubated at 30°C for 24 hours. After incubation, the strain was plated on MRS agar medium and then incubated during 48 hours and one typical colony was used to inoculate 5 mL of MRS liquid medium. After incubation at 30°C for 24 hours, the microbial suspension was used to evaluate probiotic abilities including antibacterial activity, titratable acidity, acid tolerance, proteolytic power, lipolytic power and resistance to bile salts.

2.2 Antibacterial activity of lactic acid bacteria

The evaluation of antimicrobial activity of lactic bacteria was carried out according to to the well diffusion method previously described by Tadesse et al. [15].

To study antimicrobial activity of lactic bacteria on potential pathogenic microbial, 267 lactic acid bacteria were tested. To prepare the inoculum, each strain was cultured on MRS agar medium and incubated in aerobic conditions at 37°C for 24 h. Then, a typical colony was transferred to 5.0 mL of MRS broth and incubated under aerobic conditions at 37°C for 24 h. After incubation, the resulting culture was centrifuged and the supernatant was used for antimicrobial activity testing.

Concerning the pathogen strains, APEC and Salmonella strains were previously isolated from poultry feeds in farms of Abidjan district (4). These strains cause generally the colibacillosis and salmonellosis respectively in poultry during farming. Salmonella and APEC strains were separately cultured on nutrient liquid medium and incubated during 24 hours at 37°C in aerobic conditions. After incubation, 200µL of this culture was mixed with 20 mL Mueller-Hinton agar maintained at 45°C. After homogenization, the medium was cooled in Petri dishes. After the solidification, wells were made aseptically in this MH agar (BioRad, France) with the sterile end of a Pasteur pipette and each well was filled with 100µL of the tested lactic acid bacteria preculture. The plate was refrigerated at 4°C for 2 hours to allow better diffusion of the active substance before incubation at 37°C for 24-48h. The inhibition of pathogen growth was determined by measuring the zones of inhibition and antimicrobial activity was classified according to method of Bahri [16]: (-) no inhibition; (+) weak inhibition for diameter between 0 and 3 mm; (+ +) good inhibition for diameter between 3 and 6 mm; (+ + +) strong inhibition for diameter greater than 6 mm. At the end of this test, strains with a strong antibacterial activity were selected for the further tests.

2.3 Evaluation of acidification capacity of lactic acid bacteria

Evaluation of acidification capacity of lactic acid bacteria strains was performed in MRS broth medium. Thus, three colonies of each tested strain was transferred to 10.0 mL of MRS broth and incubated under aerobic conditions at 37°C. For each strain, three tubes containing the MRS broth were inoculated. After incubation at 24 h, 48 and 72 h, one tube was took and the acid production capacity was evaluated by titration with 0.1 N NaOH solution using phenolphthalein as pH indicator according to AOAC method [17].

2.4 Acidity tolerance

To assess survival of each lactic acid bacteria at different initial culture pH values, the MRS broth (20 mL) was prepared, and the pH adjusted using acetic acid (Merck, Germany) to give a range of initial pH values from 2 to 4. Two (2) mL of each medium was inoculated with 100 µl, in triplicate, of the tested strain and incubated at 37°C during 2 hours according to the method described by Hydrominus et al. [18]. This test allows evaluating the resistance capacity of these bacteria to gastric acidity. The tolerance of acidy was evaluated by colony count at 0h and 2h on MRS agar medium and plates were incubated for 48 h at 37°C and colonies were enumerated. A control (pH 6.8) was carried out under the same conditions. The survival rate after the acid effect was evaluated compared to the control culture by using the following relation.

2.5 Resistance to bile salts of lactic acid bacteria

The methods of Ourtirane [19] were used to study the effects of bile salts on lactic acid bacteria. Thus, MRS agar was prepared and bile salt (Conda, Madrid, Spain) was added at different concentrations (0 %, 0.3%, 0.5%, 0.8%, 1 %, 1.2%, 1.5%, 1.8%, 2%) before sterilization in autoclave at 121°C for 15 minutes. Each sterilized medium was poured into petri dishes. After solidification and drying, the medium was inoculated with 0.1 ml of the tested strain and incubated at 37°C for 48 hours. After incubation viable organisms were counted and the survival rate was expressed in Log CFU by using the following relation. All tests were carried out in triplicate.

2.6 Proteolytic enzymes production capacity of lactic acid bacteria

The screening of lactic acid bacteria strains with proteolitic activity was performed in Modified MRS agar medium containing 0.25 % of glucose. After sterilization, the medium was supplemented with 10 % skim milk was added as carbon source. Inoculation of the isolates was carried out in four (4) wells 0.5 cm in diameter and 3 mm deep made aseptically in the agar. All plates were incubated at 30 °C during 48 h. After incubation, the clear zones around the wells, indicating proteolitic activity were revealed with a solution of iodine and potassium iodide (5 g potassium iodide + 1 g iodine + 330 mL distilled water) as described by Soares et al. [20].

2.7 Lipolytic enzymes production capacity of lactic acid bacteria

The lipolytic activity was evaluated on modified-MRS solid medium containing glucose (1%) and palm oil (1 %) as sole carbon source [21]. After sterilization of the medium, 7 µL of bacterial strains were cultured by wells method as previously described and incubated at 37 °C for 48 hours. The lipolytic activity was monitored by the presence of opaque zone around the wells after incubation [21].

2.8 Statistical analysis

All tests were performed in triplicate and the results were expressed as a mean ± standard deviation. The analysis of variance (ANOVA) was performed using SPSS Statistics 20 software. Duncan's 95% cut-off test was used to determine significant differences between the means.

3. RESULTS AND DISCUSSION

3. 1 Antibacterial activity of lactic acid bacteria

A total of 267 strains were tested for analysis of the antibacterial activity against the growth capacity of Salmonella and E. coli isolates. Among of them, 134 strains have induced the *Salmonella* and *E. coli* growth inhibition revealed by presence of inhibition zone around the well (Figure1). The inhibition diameters ranged from 3 to 20 mm for these 137 strains. Based on these values, the tested isolates with antibacterial activity were classified into 3 groups. The first group with low activity was 55 strains with an inhibition halo ranged from 3 and 6 mm. The second group included 54 isolates with average antibacterial activity and diameters between 6 and 8 mm. The third group included 25 strains with strong antibacterial activity and inhibition diameters greater than 8 mm (Table 1). These 25 strains were selected for the further tests.

This inhibition activity would be induced by the secretion of several bactericidal compounds produced by lactic bacteria such as organic acids (mainly lactic acid), hydrogen peroxide, diacetyl or even antibacterial substances of a natural protein calling bacteriocins [22, 23]. Gopal et al. [24] have demonstrated a synergistic action between antimicrobial protein substances and organic acids to explain the inhibitory action of probiotic bacteria.

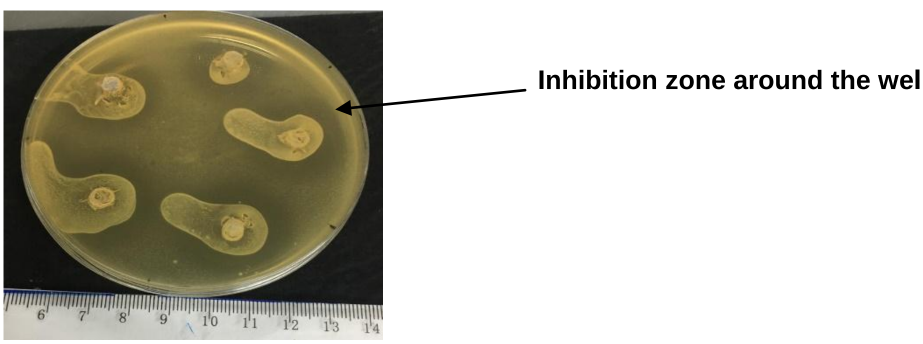

Figure 1. Growth inhibition of E. coli strains by lactic acid bacteria strain

3.2 Acid production capacity of lactic acid bacteria

The acid production capacity of lactic acid bacteria strains with high antibacterial activity was evaluated in liquid medium. The results show that the all 25 isolates previously selected were able to product high amount of acid with values ranged from 11,85 ± 1,05 % to 26,70 ± 1,20 % (Table 1). Moreover, the culture medium pH values recorded for this acidity ranged between 3.845 ± 0.08 and 4.775 ± 0.06 (Table 1).

Table 1. Titratable acidity and inhibition diameter of selected strains

Strains	Species	Titrable acidity (%)	Inhibition zone diameters (mm)

T9AB5	*Lactobacillus plantarum*	$2.37^{cd} \pm 0.90$	$17^{bcd} \pm 2.00$
T2AB1	*Lactobacillus plantarum*	$2.43^{bc} \pm 0.30$	$20^{b} \pm 0.00$
T1GB8	*Lactobacillus plantarum*	$2.25^{de} \pm 1.50$	$15^{cde} \pm 0.00$
T9AB6	*Lactobacillus plantarum*	$2.055^{f} \pm 0.45$	$16^{cd} \pm 5.00$
T0AB7	*Leuconostoc mesenteroides*	$2.445^{bc} \pm 0.15$	$17^{bcd} \pm 2.00$
T11C5	*Lactobacillus plantarum*	$2.43^{bc} \pm 2.10$	$20^{b} \pm 1.00$
T11AB16	*Lactobacillus plantarum*	$2.415^{bc} \pm 1.35$	$14^{def} \pm 2.00$
T0AB9	*Leuconostoc mesenteroides*	$2.49^{bc} \pm 0.30$	$27^{a} \pm 2.00$
T1AG22	*Lactobacillus plantarum*	$1.245^{lm} \pm 0.45$	$15^{cde} \pm 3.00$
T7C8	*Lactobacillus plantarum*	$2.565^{ab} \pm 0.75$	$11^{fg} \pm 0.00$
T0AB1	*Lactobacillus plantarum*	$2.175^{ef} \pm 1.05$	$9^{g} \pm 1.00$
T8AB5	*Lactobacillus plantarum*	$2.67^{a} \pm 1.20$	$9^{g} \pm 2.00$
T11AB17	*Lactobacillus plantarum*	$2.415^{bc} \pm 0.15$	$17^{bcd} \pm 3.00$
T7AB3	*Lactobacillus plantarum*	$1.89^{g} \pm 1.80$	$11^{fg} \pm 0.00$
LAB222	*Lactobacillus plantarum*	$1.485^{ijk} \pm 0.15$	$20^{b} \pm 1.00$
LAB115	*Lactobacillus plantarum*	$1.47^{jk} \pm 0.30$	$9^{g} \pm 1.00$
LAB65	*Lactobacillus plantarum*	$1.65^{hi} \pm 0.30$	$18^{bc} \pm 1.00$
LAB127	*Lactobacillus plantarum*	$1.665^{h} \pm 0.15$	$12^{efg} \pm 0.00$
LAB19	*Lactobacillus plantarum*	$1.74^{gh} \pm 0.60$	$15^{cde} \pm 1.00$
LAB18	*Enterococcus faecium*	$1.635^{hij} \pm 1.35$	$14^{def} \pm 0.00$
LAB26	*Lactobacillus plantarum*	$1.47^{jk} \pm 0.30$	$17^{bcd} \pm 0.00$
LAB126	*Lactobacillus plantarum*	$1.59^{hij} \pm 0.90$	$20^{b} \pm 1.00$
LAB182	*Lactobacillus plantarum*	$1.185^{m} \pm 1.05$	$17^{bcd} \pm 1.00$
LAB85	*Lactobacillus plantarum*	$1.365^{kl} \pm 0.75$	$10^{g} \pm 2.00$

3.3 Acid tolerance of lactic acid bacteria

The influence of pH on grow capacity of the selected strains indicates that these 25 LAB strains, with antimicrobial activity are also able to grow at pH 3.5 to pH 6.8 the two hours incubation time. However, the results indicate a progressive decrease of bacterial load with the decrease of pH of the culture medium and no growth was observed for the pH under 3.5.

Among the tested strains, 20 isolates were able to grow at pH ≥ 4 with high survival rates (Table 2) and six strains were able to grow at pH 3.5 with survival rate range from 30 % to 89%. In addition, *Lactobacillus plantarum* specie showed high resistance to acid shok than other species tested in this study including *Enterococcus faecium* and *Leuconostoc mesenteroides.*

In addition the tested Lactic acid bacteria strains showed excellent resistance after 2 hours at pH 4 for the all strains and at pH 3.5 for 6 isolates. In fact, the number of viable cells after 2 hours of incubation remains significant with a survival rate of more than 50 % at pH 4 and 30 % at pH 3.5 indicating a good resistance to acid stress of these strains.

Thus, the lactic acid bacteria tested strains in this study would be survive and adapt in the poultry digestive tract mainly because of pH values ranged generally between 4.47 to 6.58 from the Jabot to the colon [25, 26]. According to Gabriel et al. [27] it is in the jabot, that we mainly find lactobacilli which are attached to the epithelium and form almost a continuous layer.

3.4 Proteolytic activity of lactic acid bacteria

A total of twenty (20) strains were tested for proteolitic activity evaluation. Among them, 10 strains exibited high proteolitic activity with clear halo diameters ranging from 25 ± 3.00 to 28.67 ± 2.51 mm while 15 strains were low producers with production zone diameter under 25 mm (19 to 24.67 mm). As, a strain with the lysis zone diameter after incubation is above 5 mm is considering to have proteolytic activity, all tested strains with production diameter above 19 mm are high producer. Moreover, *Lactobacillus plantarum* (T7C8) and *Leuconostoc mesenteroides* (T0AB9) showed better proteolytic activity (Table 3).
Results of proteolyic enzymes production evaluation indicated that ten (10) tested strains exhibit high proteolyic activity and 15 isolates were considering low producer respectively with clear halo diameters ranged from 25 to 2 9 mm and 19 to 24 mm. According to Vuillemard [28], with halo diameter higher 5 mm, tested strain is considering to be proteolytic enzyme production capacity. Thus, our strains with halo diameter ranged from 19 to 29 mm, were found to be highly proteolytic. These results are in agreement with those obtained by Ayadi et al. [29]. These authors found fairly significant protease activity in *Lactobacillus* and *Leuconostoc.* In our study, the tested strains synthesized caseinase enzymes to digest milk proteins in order to use them as a substrate facilitating their growth. Thus, using of these strains as probiotics in poultry farming will allow to break down complex proteins containing in animal feeds into simple amino acids and this could improve the poultry zootechnical performance.

Table 2. Acid stress survival rate of tested strains

Strains	Species	pH 4	pH 3.5
T11AB17	*Lactobacillus plantarum*	65.19± 7.31	0
T7C8	*Lactobacillus plantarum*	61.84± 13.61	0
T7AB3	*Lactobacillus plantarum*	75.04± 4.11	0
T11AB16	*Lactobacillus plantarum*	78.93±5.3	0
T9AB6	*Lactobacillus plantarum*	68.20± 3.16	0
T0AB9	*Leuconostoc mesenteroides*	97.52± 1.34	67.23 ±3.22
T0AB1	*Lactobacillus plantarum*	67.13± 2.52	0
T9AB5	*Lactobacillus plantarum*	68.56± 3.07	0
T2AB1	*Lactobacillus plantarum*	76.75± 8.65	0
T1GB8	*Lactobacillus plantarum*	98.20± 3.21	52.33±3.45
T0AB7	*Leuconostoc mesenteroides*	72.56± 3.62	0
T11C5	*Lactobacillus plantarum*	70.11± 2.57	0
LAB 26	*Lactobacillus plantarum*	64.22± 7.73	45.25±2.11
LAB 127	*Lactobacillus plantarum*	90.66± 9.28	88.62±1.23
LAB 222	*Lactobacillus plantarum*	58.14± 3.45	0
LAB 126	*Lactobacillus plantarum*	53.35 ± 7.15	0
LAB 18	*Enterococcus faecium*	95.72± 5.05	30.21 ±4.05
LAB 65	*Lactobacillus plantarum*	48.50± 3.80	0
LAB 19	*Lactobacillus plantarum*	63.69 ± 5.82	0
LAB 182	*Lactobacillus plantarum*	57.03± 3.48	0

Table 3. Proteolytic enzymes production zone diameters of tested strains

Strains	**Species**	**Production zone diameters (mm)**
T1GB8	*Lactobacillus plantarum*	19 ± 4.24
LAB26	*Lactobacillus plantarum*	21 ± 1.41
LAB127	*Lactobacillus plantarum*	22 ± 1.41
T9AB6	*Lactobacillus plantarum*	23 ± 2.82
T11AB16	*Lactobacillus plantarum*	23 ± 0.70
LAB18	*Enterococcus faecium*	23 ± 2.82
T9AB5	*Lactobacillus plantarum*	24 ± 1.41
T7AB3	*Lactobacillus plantarum*	25 ± 2.82
T11C5	*Lactobacillus plantarum*	25 ± 1.41
LAB 182	*Lactobacillus plantarum*	25± 1.41
LAB 19	*Lactobacillus plantarum*	25 ± 1.41
T0AB7	*Leuconostoc mesenteroides*	26 ± 7.07
T11AB17	*Lactobacillus plantarum*	26 ± 4.24
T2AB1	*Lactobacillus plantarum*	26 ± 5.65
T0AB1	*Lactobacillus plantarum*	26 ± 1.41
LAB 65	*Lactobacillus plantarum*	26± 1.41
LAB 126	*Lactobacillus plantarum*	26± 8.48
LAB222	*Lactobacillus plantarum*	27 ± 1.41
T7C8	*Lactobacillus plantarum*	29 ± 8.48
T0AB9	*Leuconostoc mesenteroides*	29 ± 4.24

3.5 Lipolytic activity of lactic acid bacteria

Among the 20 isolates were tested, 10 strains showed lipolytic enzymes production activity with halo diameters ranged between 19 to 13 mm. These ten strains were classified into two (2) groups. The first group with high activity concerns three (3) strains and including T0AB7; T0AB9 and LAB 18. The production zone diameters of these high producer strains ranged between 19 and 18 mm. The second group with low capacity concerns seven strains with zone production diameters ranged from 14 and 13 mm) (Table 4). The figure 7 shows halo around the seeded wells indicating the ability of the isolates to lypolitic enzymes production.
Among the 20 tested strains, ten were able to produce lipolytic enzymes with production zone diameter ranged to between 19 to 13 mm. these results indicate the capacity of these tens isolates to also break down the complex lipids in free fatty acids.
In generally, lactic acid bacteria are considered to be weakly lipolytic compared to other bacterial species such as *Pseudomonas*, *Acetobacter* or *Flavobacterium* [30, 31, 32]. However, Karam et al. [21] have suggested that the presence of lactic acid bacteria in high concentration in cheeses and under favorable conditions can lead to the production of a significant amount of free fatty acids probably due to an adaptation to these conditions.
These strains with lipolytic activity are able to synthese extracellular lipases which convert lipids into fatty acids revealed by the presence of the opaque zone around the well.
Thus, these ten strains could be use as probiotic to improve the zootechnical performance of poultry during the farming because of available of essential fatty acids.

Table 4. Lipolytic enzymes production zone diameters of tested strains

Tested strains	Species	Inhibition zone diameters (mm)
T0AB7	*Leuconostoc mesenteroides*	19 ± 2.12
T0AB9	*Leuconostoc mesenteroides*	18 ± 2.12
LAB18	*Enterococcus faecium*	18±1.41
T9AB5	*Lactobacillus plantarum*	14±1.41
T1GB8	*Lactobacillus plantarum*	14± 1.41
T7AB3	*Lactobacillus plantarum*	14± 1.41
LAB19	*Lactobacillus plantarum*	14± 4.24
LAB127	*Lactobacillus plantarum*	14± 1.41
LAB26	*Lactobacillus plantarum*	13± 2.82
T9AB6	*Lactobacillus plantarum*	13± 1.41

3.6 Resistance of lactic acid bacteria strains to bile salts

A total of twenty (20) strains were tested to evaluate their resistance capacity at different concentrations of bile salts. Among these isolates, 14 were able to growth in presence of 0.3 % to 0.8 % of bile salts and only 6 strains were shown capacity to growth with 1 % of bile salts in the medium. These strains with high

resistance capacity to bile salts including four (4) strains of *Lactobacillus plantarum* (T1GB8, T11AB17, LAB26, LAB 127), one strain of *Leuconostoc mesenteroides* (T0AB9) and one isolate of *Enterococcus facium* (LAB18). The survival rates of these strains ranged from 15.97 to 37.41 % at 0.3 % of bile salts concentration and from 2.97 to 16.99% with 1% of bile salt in medium.
In addition, six of these tested strains exhibited a good resistance to stress of gastrointestinal tract caused by bile salts because of their growth capacity in presence of 1% of bile salts in the medium.
These strains could be very interesting as a probiotic insofar as the stress due to bile salts which varies between 0.6 and 0.8% in chickens will have no effect on their activities.

4. CONCLUSION

The lactic acid bacteria strains tested in this study show high antimicrobial activity. They also show capacity to support the intestine stress conditions. Moreover, some isolates were able to produce proteolytic and lipolytic enzymes. This study shows the possibility of use probiotics lactic acids bacteria as antibiotics alternative in poultry sector to reduce some avian pathologies affecting the poultry sector in Côte d'Ivoire.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. authors EEA sand CSD designed the study and performed the statistical analysis, author BGG wrote the protocol and the first draft of the manuscript and managed the analyses of the study. authors OHD, ls and DYN managed the literature searches. all authors read and approved the final manuscript.

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