Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 10, Issue, 05, pp.69341-69348, May, 2018

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

DIFFERENTS CHARACTERIZATION OF LACTIC ACID BACTERIA ISOLATED FROM ADIOPODOUME'S FARMS IN IVORY COAST

[1*]Dr. Ibourahema COULIBALY, [2]Dr. Elisée Kporou KOUASSI, [3]Dr. Elyse Amoin N'GUESSAN, [4]Dr. Ibrahim KONATE and [5]Pr. Daouda KONE

[1]University Jean Lorougnon Guédé - Research Unit in Agroforestry, Laboratory of Microbiology, Bio-Industry and Biotechnology, BP. 150 Daloa, Ivory Coast
[2]University Jean Lorougnon Guédé - Research Unit in Agroforestry, Laboratory of Biochemistry and Microbiology, Natural Substances, BP 150 Daloa, Ivory Coast
[3]University Peloforo Gon Coulibaly - Research Unit in Biochemistry and genetic, Laboratory of Biologicals Sciences, BP. 13280 Korhogo, Ivory Coast
[4]University Jean Lorougnon Guédé - Research Unit in Agroforestry, LaboratoryInteractions Host - Microorganism, Environnement and Evolution, BP. 150 Daloa, Ivory Coast.
[5]University Felix Houphouët Boigny - Research Unit in Biosciences, Laboratory of Plant Physiology,BP 582 Abidjan 22, Ivory Coast

ARTICLE INFO

Article History:
Received 26th February, 2018
Received in revised form
14th March, 2018
Accepted 06th April, 2018
Published online 30th May, 2018

Key words:

Characterization, Lactic Acid Bacteria, *Lactobacillus,* $L^{(+)}$-lactic acid, $D^{(-)}$-lactic acid, Optical isomers.

ABSTRACT

Isolation of lactic acid bacteria (LAB) from poultry suburbs farms of Adiopodoumé-Abidjan (Ivory Coast) was carried out. LAB were cultivated on lactic acid medium (MRS) and were characterized based on colonies morphology, cells morphology and biochemical tests. Lactic acid bacterial strains were isolated from soils, chicken faeces and feathers.Out of fifteen samples analysed, thirteen (86.6%) harboured LAB. All isolates were obtained by sequential screening for catalase activity and Gram-staining.Out of (86,6%) which five (33,3%) were established to be homofermentative by the gel plug test. Four isolates (26,6%) were identified by use of the API 50CHL kit and three*Lactobacilli* strains and one *Lactococci* (0,066%) strain were selected to study their growth and lactic acid production profiles in a time course experiment. The *Lactobacilli* strains, both isolated from faeces and fresh intestine, produced higher amounts of cells and lactic acid from soils as compared to the*Lactococci* strain isolated from feathers. $L^{(+)}$-lactic acid is the only optical isomer for use in pharmaceutical and food industries because is only adapted to assimilate this form. The opticals isomers of lactic acid were examined by $L^{(+)}$ and $D^{(-)}$-lactate dehydrogenase kit. *Lactobacilli* strains produced combination of both optical isomers of lactic acid. Among them, *Lactobacillus casei* subsp. *casei* produced the low amount of $D^{(-)}$-lactic (2%). The optimum rates of glucose for lactic acid production by *Lactobacillus* strains were 180 and 120 g/L for *Lactobacillus plantarum* and *Lactobacillus paraplantarum*, respectively.

****Corresponding author:***

Citation: Dr. Ibourahema COULIBALY, Dr. Elisée Kporou KOUASSI, Dr. Elyse Amoin N'GUESSAN, Dr. Ibrahim KONATE and Pr. Daouda KONE, 2018. "Differents characterization of lactic acid bacteria isolated from adiopodoume's farms in ivory coast", *International Journal of Current Research*, 10, (05), 69341-69348.

INTRODUCTION

Lactic acid bacteria (LAB) are group of gram-positive, non spore forming, non–respiring, cocci or rods, which produce lactic acid as the major end product of the fermentation of carbohydrate (Allen *et al*., 2001). LAB are used as natural or selected starters in food fermentation in which they perform acidification due to production of lactic acid flavor.

Bacteria from the genera*Lactobacillus, Lactococcus, Leuconostoc, Pediococcus*and *Streptococcus* are the main species of LAB involved. Lactic acid bacteria (LAB) have been isolated from many fermented foods for the use as probiotics and functional food materials (Solieri *et al*., 2014). Lactic Acid Bacteria have also been used in many fermented foods, especially in non-dairy fermented products such as products from sh, meat and vegetables (Aslim *et al*., 2005).

Lactic acid bacteria are able to produce acids, hydrogen peroxide and bacteriocins and possessedgreat potential as food bio-preservatives (Rhee *et al*., 2011). There has been increase attention in the use of diverse strains of LAB as probiotics, mainly *Lacobacilli*and *Bi dobacteria*which are residents of the commensal bacteria in the gut of human and animals, showing good therapeuticfunctions (Lavanya *et al*., 2011). Isolation and screening of microorganisms from natural sources has always been the most powerful means for obtaining useful and genetically stable strains for industrially important products. Lactic acid bacteria (LAB) are important in the food and dairy industries because the lactic acid and other organic acids produced by these bacteria act as natural preservatives as well as flavourenhancers. The ability of some *Lactobacilli* to produce neurochemicalspotentially able to reach the cerebellum via the vagus nerve (Bercik *et al*., 2011)., raises the question of whether such probiotics could benefit people with mooddisorders? Certainly animal studies have shown the microbiotagut-brainsignalling can be influenced by neural, hormonal, immune and metabolic pathways and potentially affect mood, pain and cognition (Alcock *et al*., 2014 ; Borre *et al*., 2014). Another example is the ability of lactobacilli to bind to myco- toxins produced by Aspergillus species in pre- or post-harvest cereals and milk (Hamidi *et al*., 2013). When tested in humans, a statistically significant decrease in urinary concentration occurred when probiotics were consumed twice daily for five weeks (El-Nezami *et al*., 2006). Today, lactic acid bacteria (LAB) species have become an industrially important group of bacteria used for the production of fermented foods such as yoghurt, cheese and butter. They are also crucial microbes featured in many processes used to transform and preserveourfoods (Gezginc *et al*., 2015 ; Klaenhammer *et al*., 2011). LABs are widelydistributed in the natural world and various species have been used for the production of fermented milk in many countries for thousands of years in the field of food processing (Yoshida *et al*., 2010). *Lactobacilli* are found in a variety of habitats, also in the gastrointestinal (GI) tract of human. Lactic acid bacteria have a long history as GRAS (generallyregarded as safe) organisms and especially members of genus *Lactobacillus*, *Lactococcus*and *Streptococcus* are widely used in fermentation industry. Several species of genus *Lactobacillus* have been used in food products as probiotic organisms. Probiotic strains are selected for potential application on the basis of particular physiological and functional properties. This among other impacts will increase the functionality of probiotic LABs and data thereof can be used as a good basis to further manipulate LAB genes (Song *et al*., 2014). More research into their use as functional food ingredients is currentlyunderway and is expected to increase in the nearest future. There is already growing research into the attenuating effects of probiotic LABs on breast cancer cells and the likes, thusfurtherbridging the gap between the food, health and medicinesectors of the world (Chang *et al*., 2015). Lactic acid could be produced by chemical synthesis, e.g., as a by-product in the petrochemical industries, or it could be made by microbial fermentation. The fermentation process is becoming more relevant because the rawmaterials used in fermentation are renewable in contrast to petrochemicals (Kharras *et al*., 1993). Furthermore, the fermentation process could produce optically pure isomers of lactic acid. Pure isomers, $L^{(+)}$ or $D^{(-)}$-lactic acid, are more valuable than the racemic DL form because each isomer has its own applications in the cosmetics and pharmaceuticals industries.

In PLA, the ratio of $L^{(+)}$ and $D^{(-)}$-lactic acid influences the degradability of the polymers (Panesar *et al*., 2010); therefore it is easier to manufacture PLA with specific properties, e.g., degradability, if L (+) and D (-) - lactic acid are supplied separately. LAB that have industrial potential should be homofermentative, i.e., they produce only lactic acid. The lactic acid bacteria (LAB) are of major economic and nutritional importance because of their use in the production and preservation of a many variety of fer- mented products. In particular, species closelyrelated to the *Lactobacillus* group are increasingly used as adjunct cultures in milk fermentations (Narayanan *et al*., 2004). Much current research is focused on characterizing their industrially relevant traits and the opportunities for strain improvement by metabolic engineering (Wee *et al*., 2006 ; Guha *et al*., 2003). Lactic acid bacteria are characterized as Gram-positive, usually non-motile, non-sporulating bacteria that produce lactic acid as a major or sole product of fermentative metabolism. (Kandler, 1986) have classified *Lactobacillus* isolates from temperate regions according to their morphology, physiology and molecular characters (Kandler, 1986; Reid, 1999) classified LAB based on the molecular characteristics. LAB from food and their current taxonomicalstatus have been described by many (Gonzalez *et al*., 2000; Ringoe, 1998; Gasson, 1993). Taxonomic studies on LAB from African animals are rare. Molecular methods are important for bacterial identification (Drancourt *et al*., 2001 ; Greetham *et al*., 2003; Mayra-Makinen , 1998) and possibly more accurate for LAB than are conventionalphenotypicmethods. The objective of this study was to isolate lactic acid bacteria from chicken faeces and poultry soils and to screen these isolates for desirable traits such as homofermentative ability, production of isomeric form lactic acid, and tolerance to high concentration of lactic acid, low pH and high temperature.

MATERIALS AND METHODS

Isolation of lactic acid bacteria strains : Soils, fresh intestine, feathers and faeces samples were collected from different poultry farms in Adiopodoumé-Abidjan (Ivory Coast, West Africa). Strains isolation was carried out according to (Nakayama, 1967). Five grams of sample were mixed with 100 ml of GYP medium contained 1% glucose (w/v), 1% yeast and 1% peptone. The sample suspension was heated for 10 min at 80°C and incubated anaerobically at 30°C. After 48 h of incubation, 100 µl of the mixture were spread onto the surface of GYP agar containing 1% CaCO3. The culture was incubated anaerobically at 30°C. Acid producing bacteria were recognised by the clear zones around the colonies. Bacteria were purified by several isolations and fresh cultures of these isolates were conserved at -80°C with glycerol (30%) as cryoprotective agents (33). Each of the isolates was first tested for catalase by placing a drop of 3% hydrogen peroxide solution on the cells. Immediate formation of bubbles indicated the presence of catalase in the cells. Only those isolates which were catalase- negative were Gram-stained, and only those which were Gram- positive were put through the gel plug test (34) to determinewhether the isolate produced carbon dioxide during fermentation. An isolate was deemed to be a homofermentative lactic acid producer if no gas was produced. Based on the results, five homofermentative isolates were selected for further studies. These isolates were identified by the API 50CHL identification kit (BioMérieux, Marcy-l'Etoile, France).

Determination of lactic acid isomers : Bacterial cell concentration was determined at 610 nm and calibrated into colony forming units (CFU) by colony count method and into dry mass weight (Reid *et al*., 2013). Bacterial cell concentration was determined by dry mass weight (Kumar *et al*., 2012 ; Monteagudo *et al*., 1997). The dry mass weight of bacterial and fungal cells was determined by centrifugation of the fermentation broth and freeze-dried sediments (Monteagudo *et al*., 1997). The concentration of lactic acid was measured based on colorimetric determination by $L^{(+)}$ and $D^{(-)}$-lactate dehydrogenase kit (Megazyme International Ireland Ltd., Co. Wicklow, IRELAND). In this method, known amounts of production medium were taken during fermentation and centrifuged at 3000 × g for 10 min. The supernatant was used directly for determination of lactic acid concentration. The assays are specific for either D-lactic acid or L-lactic acid. In the assay of lithium D-lactate (MW = 96.0) results of approx. 96% (w/w) can be expected, while a value of 98% (w/w) should be obtained with lithium L-lactate. The smallest differentiating absorbance for the assay is 0.005 absorbance units. This corresponds to 0.107 mg/L of sample solution at the maximum sample volume of 1.50 mL (or to 1.60 mg/L with a sample volume of 0.1 mL). The absorbance of the solutions (A_1) was read against blank at 340 nm (Beckman Coulter, spectrometer AD 340s) after approx. 3 min and the reactions were initiated by addition of 0.02 ml of (D-LDH)/(L-LDH) suspensions and then the second absorbances of the solutions (A_2) were read at the end of the reaction (approx. 20 min). In situations where the reaction did not stop after 20 min, the absorbances were read at 5 min intervals until the absorbance either remains the same, or increases constantly over 5 min. The $D^{(-)}$ and $L^{(+)}$-lactic acid were also determined by enzyme test kit according to the manufacturer's instruction (Megazyme International Ireland Ltd).

Assay of residual glucose : Consumption of glucose during cultivation is measured at the end of fermentation. The diluted supernatant as above was also used for the assay of residual glucose. After centrifugation, the supernatant was collected for analysis. The determination of glucose in the supernatant was measured by using an analyzer of sugar, YSI2700 SELECT (Yellow Springs Instrument Co., Inc.).

Effect of temperature, pH, lactic acid and sodium chloride : A basal MRS medium was used in these series of studies (De Man, 1960) but without beefextract, and with 0.15 g/l bromocresolpurpleadded as pH indicator (pH 7). A lowering of this pH would change the medium from purple to yellow, and was taken to indicate cell growth because the production of lactic acid is growth- related. No change in the colour of the medium was taken to indicate no cell growth because no acids were released. Universal bottles with screw caps were each filled with 20 ml of the basal MRS medium and autoclaved. An 18 h culture of each isolate was used as the inoculum whereby the cells were spin down, resuspended in 0.85% saline, and 100 µl of the suspension was inoculated into each test bottle. The temperatures tested were 15, 37 and 55°C, the concentrations of lactic acid tested were 3, 7 and 12% (w/v), and the concentrations of NaCl tested were 1.5, 5and 10% (w/v). Three pH were tested, i.e., 4,7 and 9. The basal MRS medium was adjusted with 1 M phosphoric acid and 1 M NaOH to prepare this initial pH. The bottles were placed in water baths with reciprocal shaking, set at the specific test temperatures or at 37°C for the tests on pH and concentrations of lactic acid and NaCl.

At the end of 42 h, the colour changes and turbidity of each bottle was noted as a simple indication of growth or no-growth. Each treatment was tested with triplicate bottles.

Determination of turbidity : The biomass evolution was followed by using the turbidimetrical method (the optical density : OD) at 540 nm. Acidity of the culture media was measured by titration with 0.5 N NaOH using phenolphthalein as pH indicator.

RESULTS AND DISCUSSION

Screening of lactic acid bacteria : Thirty bacterial isolates were obtained from the poultry farms in Ivory Coast (Adiopodoumé-Abidjan) and glucose was used in the enrichment medium. Of these, 25 tested Gram-positive and catalase-negative, and all but one was rod-shaped. The one remaining LAB isolate was coccus-shaped. Fifteen of these isolates did not produce gas in the gel plug test and were, therefore, deemed to be homofermenters. Based on the origin which the isolate was obtained, four of these 15 were selected for characterization and the tolerance tests. Table 1, lists these four isolates, giving details about the matrice origin, according to the substrate used in the enrichment process (sugar), cell morphology, and identification by the API 50CHL kit, probability of fit to the closest species, and the type of lactic acid isomer produced. The isolate from feathers regardless of the sugar used in the enrichment medium, were identified as *Lactobacillus casei*, while the isolate from soils was identified as *Lactobacillus plantarum*. The isolate obtained from fresh intestine was identified as *Lactobacillus lactis*. Isolates identified as *L. casei* and *L. lactis* produced only the L-form of the lactic acid while the isolate identified as *L. paraplantarum* produced a mixture of D and L isomers of lactic acid. LAB strains that produce a single isomeric form of lactic acid are more desirable as industrial strains compared to those strains that produce a racemic mixture of lactic acid. This is because the material properties of poly (lactic acid) are governed by the ratio of L and D isomers in the polymer, so the polymerization process would be better controlled when the lactic acid used as starting material is in single isomeric forms.

Tolerance to high temperatures, concentrations of lactic acid and sodium chloride, and low pH : Table 2 shows the tolerance of each of the five selected isolates to the environmental conditions tested. Three strains of the *Lactobacillus* sp. grew at 15 to 37°C, while the isolated from feathers, Abj1, could not grow at 15°C. The *Lactococcus*, Abj2, isolated from fresh chiken's intestine grew at higher temperatures ranging from 30 to above 37°C with no growth indicated at 15°C. (39) noted reducedglycolytic activity leading to reduced production of lactic acid in *L. lactis* at low temperature. The ability to grow at high temperature is a desirable trait as it could translate to increased rate of growth and lactic acid production. At the same time, a high fermentation temperature reduces contamination by other microorganisms. Abj2, however, was the least tolerant to high concentrations of lactic acid as growth was indicated only at 2 and not at 5% and higher. The two isolates from soils and chicken faeces, Abj3 and Abj4 respectively, were the most tolerant of high lactic acid concentrations as they were able to grow at 7%. None of the fourisolates grew above 7% lactic acid concentrations. A higher tolerance to lactic acid is a desirable trait for an industrial strain of LAB as it could

Table 1. Characteristics of four homofermentative LAB, isolated from poultry frams in Adiopodoumé (Abidjan's suburbs)

Isolate name	Origin of isolates	Cell morphology	Identification API 50 CHL	Identification probabilities (%)	LAB isomer
AbjA1	Feachers	Rod	*Lactobacillus casei*	97,7	L(+)
AbjA2	Fresh intestine	Cocci	*Lactobacillus lactis*	98,6	L(+)
AbjA3	Soils	Rod	*Lactobacillus plantarum*	99,8	D/L
AbjA4	Chicken faeces	Rod	*Lactobacillus paraplantarum*	99,7	D/L

Table 2. Tolerance of four LAB isolates to ranges of temperatures, lactic acid, NaCl concentrations and pH

Culture conditions	Strains names			
	AbjA1	AbjA2	AbjA3	AbjA4
Temperature				
15°C	-	-	+	+
37°C	+	+	+	+
55°C	-	-	-	-
Lactic Acid Concentration (%, W/V)				
3.0	+	+	+	+
7.5	-	-	+	+
12.0	-	-	-	-
NaCl Concentration (%, W/V)				
1.5	+	+	+	+
5.0	+	+	+	+
10 .0	-	-	-	-
pH (End point)				
4.0	+	-	+	+
7.0	+	+	+	+
9.0	+	+	+	-

+ indicates colour change from purple to yellow, taken to equate growth ;
-indicates colour no change from purple, taken to equate no growth.

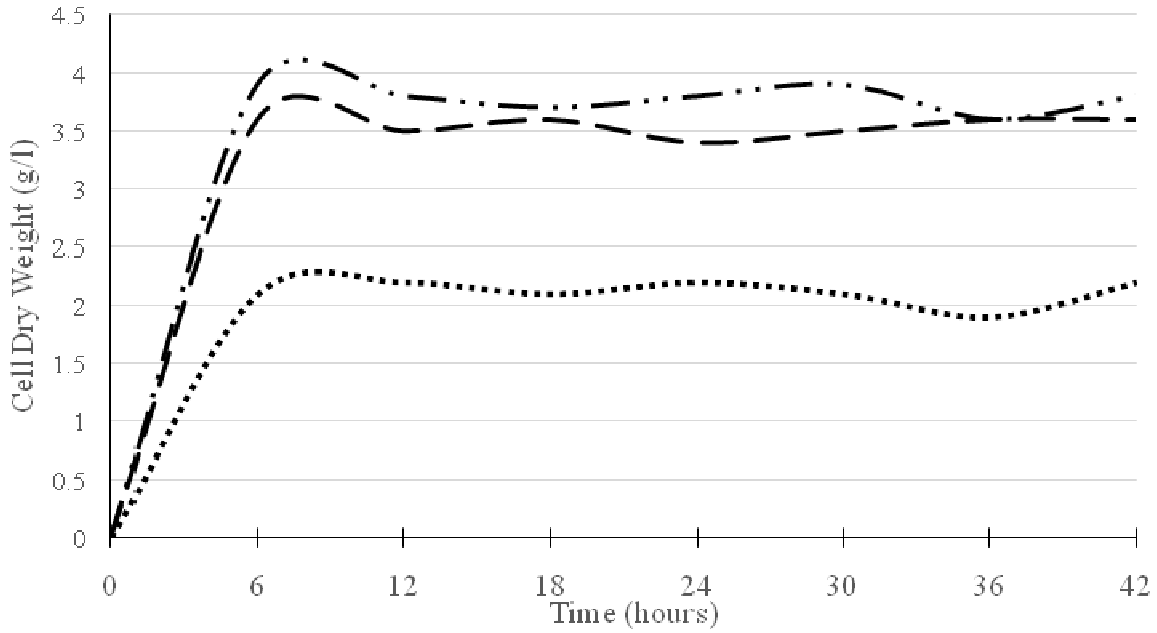

Figure 1. Biomass produced by three strains *Lactobacilli :* : (●●●) Abj2, (—●●—) Abj3 and (—) Abj4 isolates over 42 h.

produce more lactic acid in the fermentation broth without prematurely affecting itself adversely. In contrast to its low tolerance to lactic acid, Abj2 was the most tolerant to high NaCl concentration compared to the other isolates. Abj2 grew in concentrations up to 7% while the rest could grow up to 5%. None of the isolates could grow in 5% NaCl.

This test gave an indication of the osmotolerance level of a LAB strain. Bacterial cells cultivated in a high salt concentration would experience a loss of turgor pressure, which would then affect the physiology, enzyme activity, water activity and metabolism of the cells (Sanders, 2001).

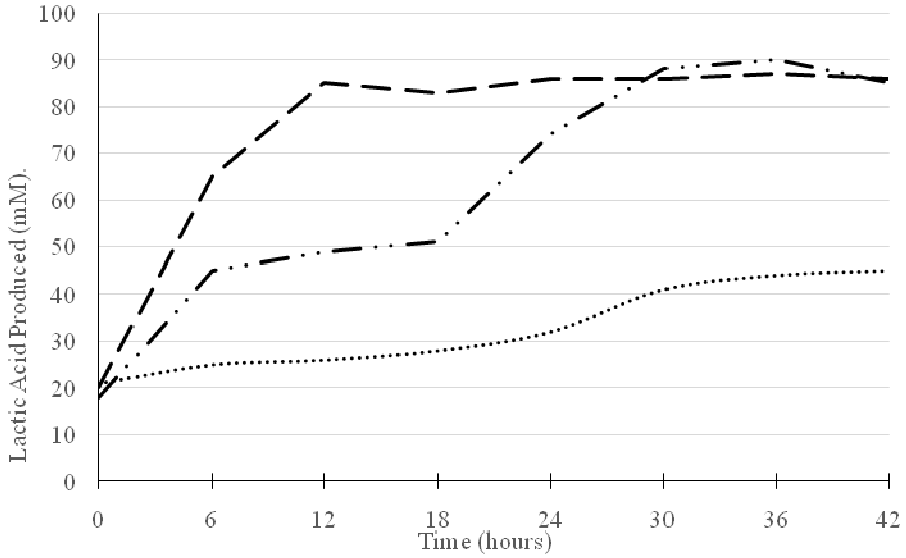

Figure 2. Lactic acid produced by three strains *Lactobacilli :* : (•••) Abj2, (—••—) Abj3 and (—) Abj4 isolates over 42 h.

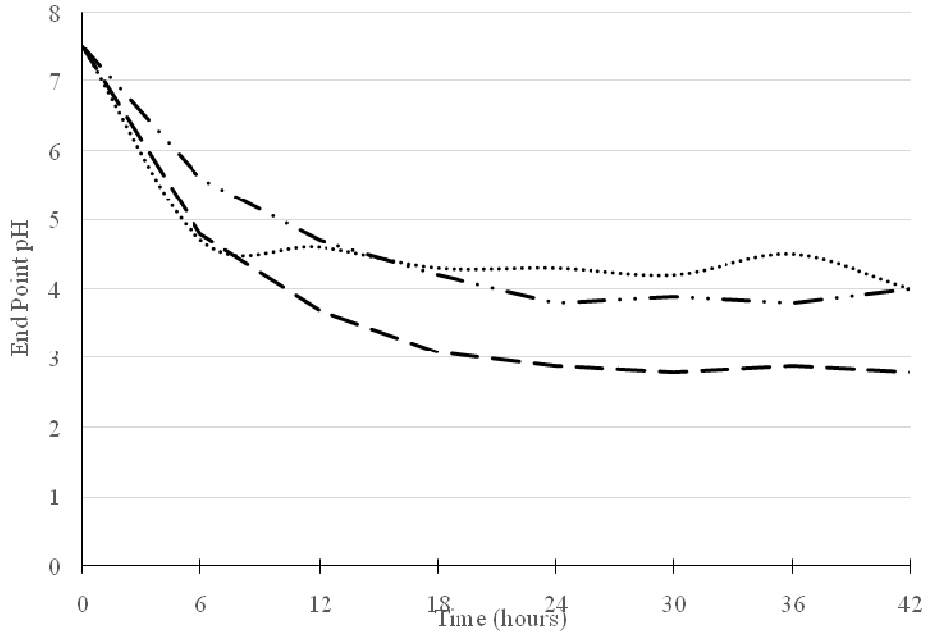

Figure 3. pH of the cultures of three strains of *Lactobacilli :* : (•••) Abj2, (—••—) Abj3 and (—) Abj4 isolates over 42 h.

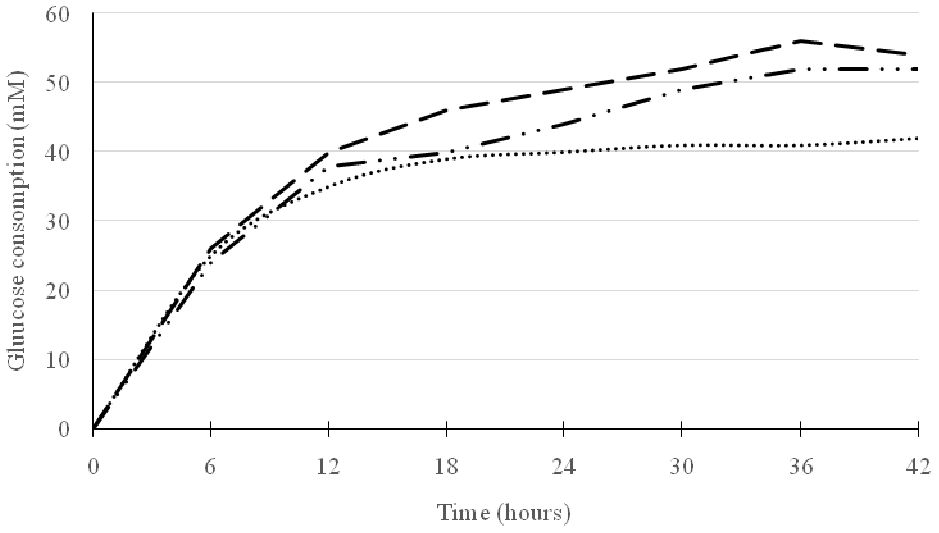

Figure 4. Glucose consumption by three strains of *Lactobacilli* : (•••) Abj2, (—••—) Abj3 and (—) Abj4 isolates over 42 h.

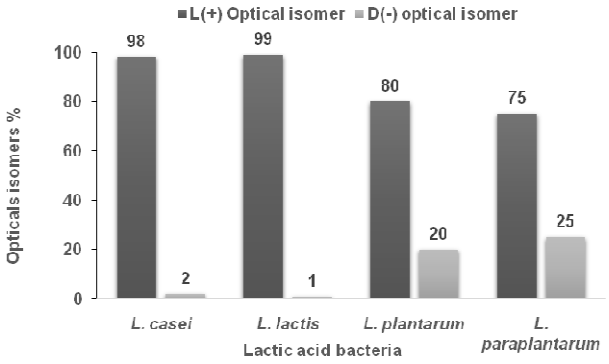

Figure 5. The results of optimum isomers of lactic acid produced by best strains

Some cells overcome this effect by regulating the osmotic pressure between the inside and outside of the cell (Kashket, 1987).There are reports describing strains of *Lactococci* (Bujnakova, 2014) and *Lactobacilli* (Hutkins *et al*., 1987 ; Glaasker *et al*., 1998)showing decreased growth rate with increasing osmolarity of the medium (Bujnakova, 2012)also noted increased amount of glycine betaine, an osmolyte, in lactococci cells when they were grown in high NaCl concentration.(40)reported that the uptake of glycine betaine was induced in cells as an adaptive measure to withstand increasing external osmotic pressure. The *Lactococci* isolate, Abj2, could be similarly protected to be able to grow at higher NaCl concentration compared to the other isolates which were lactobacilli. During industrial fermentation, as lactic acid is being produced by the cells, alkali would be pumped into the broth to prevent excessive reduction in pH. Thus, the free acid would be converted to its salt form which would in turn increase the osmotic pressure on the cells. Therefore, a LAB strain with high osmotolerance would be desirable as an industrial strain. All the isolates, except Abj2, could grow at pH 5.5. The inability of Abj2 to grow at low pH was consistent with its inability to grow at high lactic acid concentration, and was attributed to the low tolerance of *L. lactis* to free acid (H^+) compared to *Lactobacillus* spp.(41). LAB are acidophilic but while that means a tolerance to low pH, the latter should be differentiated from a situation in which a high concentration of free acids (H^+) also exists because the free acids could inhibit growth (Hutkins, 1987). On the other hand, all the isolates except Abj4, lactobacilli, could grow in alkaline environment, pH 9 (Mollet, 1999) observed that *Lactobacillus bulgaricus* also could not tolerate high pH.

Growth and lactic acid production profiles : In the first 4 h, Abj2, the lactococci strain, grew faster than Abj3 and Abj4, the *Lactobacilli* strains, based on cell dry weight (cdw) measurements. After this, the growth of Abj2 leveled off at about 1.8 g/l cdw while Abj3 and Abj4 continued to register biomass increase until 19 h before their growth too leveled off at around 3.4 and 2.8 g/l cdw, respectively (1). The low biomass produced by Abj2 with respect to Abj3 and Abj4 correlated well with the lower amount of lactic acid produced by Abj2 (Figure 5) resulting in a higher pH in the medium (Figure 2) and lower consumption of glucose (Figure 4). These results were consistent with those shown in Table 2 where Abj2 was found to be unable to tolerate high lactic acid concentrations and low pH compared to Abj3 and Abj4. While the growth profiles of Abj3 and Abj4 did not appear to be different from one another (1), the amount of lactic acid produced by Abj3 was higher than Abj4 for the first day, 24 h (Figure 5). The production of lactic acid by Abj4 also peaked faster at 18 h compared to glucose consumption (Figure 4) of the two *Lactobacilli* strains, respectively. At 30 h, the yield of lactic acid from glucose was highest with Abj3 at 1.9, followed by Abj4 at 1.6, and Abj2 at 1.5. This indicated that Abj3 had the highest efficiency in converting glucose to lactic acid. Abj3 and Abj4 had similar tolerance levels to high temperature (up to 42°C), lactic acid concentration (7.5%), NaCl concentration (5%), and to low pH (5.5), but the faster production of lactic acid by Abj4 might give it a slight advantage over Abj3. This difference between Abj3 and Abj4 was apparent under current experimental conditions, i.e., using MRS medium in which glucose was the primary carbon source and the medium was not pH-controlled. In developing the fermentation process to industrial level, cheaper sources of carbon are necessary and the medium would need to be pH- controlled by incorporating neutralizing agents such as calcium carbonate into the medium to reduce the inhibitory effects of free lactic acid on the producer cells. Under such conditions, the growth and lactic acid production of Abj3 and Abj4 would need to be re-evaluated. The time-course study was conducted to compare the growth and lactic acid producing capacity of two lactobacilli strains and a lactococci strain, the findings of which supported the results of the rapid screening tests on tolerance of the strains to a range of environmental factors. There was little doubt that the amount of biomass and lactic acid produced by the respective strains were limited by the accumulation of lactic acid in the fermentation broth and the prevailing pH, and reflected the different tolerance levels of the strains. A pH-controlled medium would probably allow for higher accumulation of lactic acid and would be appropriate for future studies where optimization of the fermentation process was the focus.

Optical isomer of lactic acid comparison: $L^{(+)}$-Lactic acid is more important for pharmaceutical and food industries; therefore produced lactic acid was used for optical isomers determination. This was examined by lactate dehydrogenase kit enzyme test. The results of optimum isomers of lactic acid produced by best strains are shown in Figure 5. The basic difference between this group *L. casei, L. lactis* and the other *L. plantraum and L. paraplantarum* fermentation is that only the form $L^{(+)}$-lactic acid is produced, whereas the latter fermentation is anaerobic and $L^{(+)}$-, $D^{(-)}$-, DL-lactate is produced. The purity of monomers is highly critical in the synthesis of polylactides and a purity of 99% or greater is usually required with the starting lactide material (Lewis, 1991). Among *Lactobacilli* strains, *L. casei* subsp.*casei* produced high concentration of $L^{(+)}$-lactic acid with 98% purity. Other *Lactobacilli* strains produced combination of both optical isomers. This confirmed experimental work of(48). Lactic acid molecule has two optical active isomers, D(-) and L(+) forms (Senthuran *et al*., 1997). Lactic acid is an organic acid with a wide variety of industrial applications. The most important application as a preservative and acidulant in foods (Senthuran *et al*., 1997), as a prostheticdevice, controlled delivery of drugs in pharmaceutical agents, as a precursor for production of polymers like polylactic acid (Dunn *et al*., 1988) and as a moisture agents in cosmetics (Dunn *et al*., 1988 ; Soccol *et al*., 1994).

Conclusion

This study described the sequential steps of isolating bacteria from southAfrican poultry farms (Ivory Coast), screening the isolates for LAB traits, selection of isolates based on a series of

tests for industrially-desirable traits, and finally compared the growth and lactic acid production profiles between the lactobacilli and lactococci strains. In this study, the determination of optical active isomer producers of lactic acid in the fermentation broth and provides a complete profile of substrate utilization by these microorganisms.

REFERENCES

Allen, A. E., Booth, M. G., Frisher, M. E., Verity, P. G., Zehr, J. P. and Zani, S. 2001. Diversity and detection of nitrate assimilation genes in marine bacteria. Applied and *Environmental Microbiology* 67(11): 5343-5348.

Solieri, L., Bianchi, A., Mottolese, G., Lemmetti, F. and Giudici, P. 2014. Tailoring the probiotic potential of non-starter *Lactobacillus* strains from ripened Parmigiano Reggiano cheese by in vitro screening and principal component analysis. *Food Microbiology* 38: 240–249.

Aslim, B., Yukesekdag, Z. N., Sarikaya, E. and Beyatli, Y. 2005. Determination of the bacteriocin-like substances produced by some lactic acid bacteria isolated from Turkish dairy products. *LWT-Food Science and Technology,* 38: 691-694.

Rhee, S. J., Lee, J. E. and Lee, C. H. 2011. Importance of lactic acid bacteria in Asian fermented foods. *Microbial Cell Factories*, 10(1): S5.

Lavanya, B., Sowmiya, S., Balaji, S. and Muthuvelan, B. 2011. Screening and characterization of lactic acid bacteria from fermented milk. *British Journal of Dairy Sciences,* 2(1): 5-10.

Bercik, P., Park, A. J., Sinclair, D., Khoshdel, A., Lu, J., Huang, X., *et al*. 2011. The anxiolytic effect of Bifidobacterium longum NCC3001 involves vagal pathways for gut-brain communication. *Neurogastroenterology and Motility*, 23, 1132e1139.

Alcock, J., Maley, C. C. and Aktipis, C. A. 2014. Is eatingbehaviormanipulated by the gastrointestinal microbiota? Evolutionary pressures and potential mechanisms. *Bioessays*, 36, 940e949.

Borre, Y. E., Moloney, R. D., Clarke, G., Dinan, T. G., &Cryan, J. F. 2014. The impact of microbiota on brain and behavior: mechanisms and therapeutic potential. Ad-vances in Experimental Medicine and Biology, 817, 373e403.

Song D, Ibrahim S, Hayek S. 2015. Recent application of probiotics in food and agricultural science. 2015 (cited Dec 10). DOI: 10.5772/50121.

Liu, Z., Qin, H., Yang, Z., Xia, Y., Liu, W., Yang, J., *et al*. 2011. Randomisedclinicaltrial: the effects of perioperative probiotic treatment on barrierfunction and post-operativeinfectious complications in colorectal cancer surgery - a double- blind study. *Alimentary Pharmacology and Therapy,* 33, 50e63.

Reid, M. K. E., Gough, R., Enos, M. and Reid, G. 2013. Social businesses in Tanzaniatackling health issues of the Millenium Development Goals, one communitykitchen at a time. *Journal of Social Business*, 3, 24e38.

Hamidi, A., Mirnejad, R., Yahaghi, E., Behnod, V., Mirhosseini, A., Amani, S., *et al*. 2013. The aflatoxin B1 isolating potential of two lactic acid bacteria. Asian *Pacific Journal of Tropical Biomedicine*, 3, 732e736.

El-Nezami, H. S., Polychronaki, N. N., Ma, J., Zhu, H., Ling, W., Salminen, E. K., *et al*. 2006. Probiotic supplementation reduces a biomarker for increased risk of liver cancer in young men from Southern China. *American Journal of Clinical Nutrition*, 83, 1199e1203.

Gezginc Y, Topcal F, Comertpay S, *et al*. 2015. Quantitative analysis of the lactic acid and acetylde- hyde produced by Streptococcus thermophilus and Lactobacillus bulgaricus strains isolated from tradi- tional Turkish yoghurt using HPLC. *J Diary Sci.,* 98:1426–1434.

Klaenhammer TR, de Vos WM. 2011. An incrediblescien-tificjourney. The evolutionary tale of the lactic acid bacteria. In : Ledeboer A, Hugenholtz J, Kok J, Konings W, Wouters J, editors. The 10th LAB symposium. Thirty years of research on lactic acid bacteria. Rotterdam : 24 Media Labs; pp. 1–11.

Yoshida Y, Seki T, Matsunaka H, *et al*. 2010. Clinical effects of probiotic Bifidobacterium breve supplementation in adult patients with atopic dermatitis. *Yonago Acta Medica*. 53:37–45.

Song, Q., Christiani, D. C., Wang, X. and Ren, J. 2014. The global contribution of outdoor air pollution to the incidence, prevalence, mortality and hospital admission for chronic obstructive pulmonary disease:a systematic review and meta-analysis. *International Journal of Environmental Research and Public Health*, 11, 11822e11832.

Chang CK, Wang SC, Chiu CK, *et al*. 2015. Effect of lactic acid bacteria isolated from fermented mustard on immunopotentiating activity. *Asian J Trop Biomed.,* 5(4):281–286.

Kharras GB., Sanchez-Riera F., Severson DK. 1993. Polymers of lactic acid. In:*Molby*, D.B. (Ed.), Plastics from microbes: Microbialsynthe- sis of polymers and polymer precursors. Hanser Publ., pp. 93-137.

Panesar, P.S., Kennedy, J.F., Knill, C.J., Kosseva, M. 2010. "Production of L(+) Lactic Acid using Lactobacillus casei from Whey. Braz. Arch. Biol. Technol., 53: 219-226.

Narayanan, N., Roychoudhary, P.K., Srivastava, A. 2004. L (+) Lactic acid fermentation and its product polymerization. Electronic J. Biotechnol., 7: 46-9.

Wee, Y.J., Yum, J.S., Kim, D., Ryu, H.W. 2006. Batch and repeated batch production of L (+)-lactic acid by Enterococcus faecalis RKY1 using woodhydrolyzate and corn steepliquor. *J. Ind. Microbiol. Biot*., 33: 431-35.

Guha, A., Banerjee, S., Bera, D. 2013. Production of Lactic acid from Sweetmeat industry waste by Lactobacillus delbruki. IJRET., 630-634.

Kandler O, Weiss N 1986. In: Bergey'sManual of Systematic Bacteriology. Sneath PHA, Mair NS, Sharpe ME, Holt JG (Eds), Vol. 2, Baltimore: Williams and Wilkins, pp.1209-1234.

Reid G 1999. The scientific basis for probiotic strains of *Lactobacillus*. *Appl. Environ. Microbiol*. 65: 3763-3766.

Gonzalez CJ., Encinas JP., Gracia-Lopez ML., Otero A. 2000. Characte- rization and identification of lactic acid bacteria from freshwaterfishes. *Food Microbiol*. 17 : 383-391

Ringoe E., Gatesoupe FJ., 1998. Lactic acid bacteria in fish: a review Aquaculture. 160: 177-203.

Gasson MJ 1993. Progress and potential in the biotechnology of lactic acid bacteria. FEMS. *Microbiol. Rev*., 12 : 3-20.

Drancourt M, Bollet C, Carta A, Rousselier P 2001. Phylogenetic analyses of *Klebsiella* species delineate *Klebsiella* and *Raoultella*gen. nov., with description of *Raoultellaornithinolytica* comb. nov., *Raoultellaterrigena* comb. nov. and *Raoultellaplanticola* comb. nov. Int. J. Syst. Evol. Microbiol. 51: 925-932.

Greetham H., Greetham L., Glenn R., Gibson CG., Hans H, Birgit M., Ulrike S., Enevold F., Matthew DC. 2003. *Clostridium colicanis*sp. nov., from canine faeces. *Int. J. Syst. Evol. Microbiol*. 53: 259-262.

Mayra-Makinen, A., Bigret M., 1998. Industrial use and production of lactic acid bacteria. In *Lactic Acid Bacteria.* Microbiology and Functional Aspects, 2nd. (eds)S. Salminen and A. V. Wright. New York: Marcel Dekker. pp. 73-102

Nakayama, O., Yanoshi, M. 1967. Spore-bearing lactic acid bacteria isolated from rhizosphere. I. Taxonomic studies on *Bacillus laevolac- ticus nov. sp.* and *Bacillus racemilacticus*nov. *sp*. *J. Gen. Appl. Microbiol*. 13 : 139-153.

De Vos, WM., Kleerebezem, M., Kuipers, OP. 1997. Expression systems for industrial Gram-positive bacteria with low guanine and cytosine content. Curr. Opin. Biotechnol. 8: 547-553.

Gibson T., Abdel-Malek T1945. The formation of carbon dioxide by lactic acid bacteria and Bacillus licheniformis and a cultural method of detecting the process. *J. Dairy. Res*., 14 : 35-44.

Reid, M. K. E., Gough, R., Enos, M. and Reid, G. 2013. Social businesses in Tanzaniatackling health issues of the Millenium Development Goals, one communitykitchen at a time. *Journal of Social Business*, 3, 24e38.

Kumar, A. M. and Murugalatha, N. 2012. Isolation of *Lactobacillus plantarum* from cow milk and screening the presence of sugar alcohol producing gene. *Journal of Food Microbiology and Antimicrobial* 4(1): 16-22.

Monteagudo JM., Rodriguez, L., Rincon, J., Fuertes, J. 1997. Kinetics of lactic acid fermentation by *Lactobacillus delbrueckii* grown on beet molasses. *J. Chem. Tech. Biotech*. 68 : 271-276.

De Man JC, Rogosa M, Sharpe ME 1960. Medium of *Lactobacilli*. *J. Appl. Bacteriol.* 23: 130-135.

Holzapfel, W.H., P. Haberer, R. Geisen, J. Björkroth, U. Schillinger. 2001. Taxonomy and important features of probiotic microorganisms in food nutrition. *Am. J. Clin. Nutr*. 73, 365S-373S.

Sanders ME, Klaenhammer TR 2001. Invitedreview: the scienti c basis of Lactobacillus acidophilus NCFM functionality as a probiotic. *J Dairy Sci*., 84: 319-331.

Kashket ER 1987. Bioenergetics of lactic acid bacteria : Cytoplasmic pH and osmotolerance. FEMS. *Microbiol. Rev*., 46: 233-244.

Bujnakova D., Strakova E., Kmet V., 2014. In vitro evaluation of the safety and probiotic properties of Lactobacilli isolated from chicken and calves. Anaerobe29: 118-127.

Hutkins RW, Ellefson WL, Kashket ER 1987. Betaine transport imparts osmotolerance on a strain of *Lactobacillus acidophilus*. *Appl. Environ. Microbiol.* 43 : 2275–2281

Glaasker E, Tjan FSB, Steeg PFT, Konings WN, Poolman B 1998. Physiological response of *Lactobacillus plantarum* to salt and nonelectrolyte stress. *J. Bacteriol*. 180: 4718-4723.

Amrane A, Prigent Y., 1999. Effect of the main culture parameters on the growth and production coupling of lactic acid bacteria. Appl. Microbiol. 2: 101-108.

Mollet B 1999. Genetically improved starter strains: opportunities for the dairy industry, *Int. Dairy*. J. 9: 11-15.

Lewis DH 1991. Controlled release of bioactive agents lactide/ glycolide polymers. In:*Polymers* as drugdeliverysystems. (Chasin, M. and Langer, R. eds.), Dekker Inc, New York, pp. 1-41.

Saeed, M., Sadeghi, N., Sharafi, M. F., Farzaneh, A. M., and Mohammad, R. B ., 2002. Comparison of Lactic Acid Isomers Produced by Fungal and Bacterial Strains. *Iran. Biomed. J*. 6 : 69-75

Senthuran, A., Senthuran, V., Mattiasson, B. and Kaul, R. 1997. Lactic acid fermentation in a recycle batch reactor using immoblized *Lactobacillus casei*. *Biotechnol. Bioeng*. 55 (6): 841-853.

Dunn, R.L., English, J.P., Strobel, J.D., Cowsar, D.R. and Tice, T.R. 1988. Preparation and evaluation of lactic acid/glycoside copolymers for drugdelivery. In: Polymers in medicine III. (Migliaresi, C. ed.), Elsevier, Amsterdam, pp. 149- 159.

Soccol, C.R., Stonoga, V.I. and Raimbault, M. 1994. Production of L-lactic acid by Rhizopus species. *World J. Microb. Biotech*. 10: 433-436.

****