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# ISOLATION AND IDENTIFICATION OF LACTIC ACID BACTERIA FROM COW MILK AND MILK PRODUCTS

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# **Article Info**



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#### **Abstract**

Lactic acid bacteria (LAB) play a crucial role in the fermentation and preservation of cow milk and milk products. This study aimed to isolate and identify LAB from cow milk and various milk products, determine the bacterial load and determine the sensory evaluation of LAB in the cow milk. A total of seven samples were collected from different locations at Bodinga metropolis and analysed using [methods, e.g., microbiological techniques]. The isolated LAB strains were characterized based on their cultural, morphological, microscopic view, biochemical, and genetic properties. Results revealed the presence of five LAB species with their percentage of occurrence, including Lactobacillus acidophilus (28.51%), Lactococcus lactis (28.51%), Lactobacillus bulgaricus (14.28%), Lactobacillus casei (14.28%), Lactococcus lactis (14.28%). The dominant LAB species varied among the different milk products, with Lactobacillus spp being the most prevalent in cow. Sensory characteristic and pH level of the cow milk and milk products was also measured. This study contributes to the understanding of the diversity and distribution of LAB in cow milk and milk products, highlighting their potential applications in food fermentation, preservation, and human health. The findings provide valuable insights for the development of novel functional foods and probiotics.

#### **Keywords:**

Lactic acid, Bacteria, Milk, Isolation, Cow

#### **CHAPTER ONE**

#### 1.0 INTRODUCTION

People have historically consumed lactic acid bacteria (LAB) in fermented foods such as dairy products. LAB are currently the subject of extensive international research due to their critical involvement in most fermented foods, as well as their potential to create antimicrobial chemicals that promote probiotic characteristic including antitumor activity, alleviation of lactose intolerance, a decrease of serum cholesterol, gut microflora stabilization, and stimulation of the immune system (Harrison et al., 2017). In the production of fermented milk, the LAB strain produced for exopolysaccharides is used to improve their viscosity and smoothness. Some LAB strains are known to produce the presumed effects of mannitol which promote health. Probiotics are live bacteria or yeasts that, when given in sufficient proportions, provide a health benefit to the host (Talei et al., 2023).

To date, with the growing interest in health consciousness, the concept of probiotic foods has gotten a lot of attention. A large number of probiotic species and strains belong to the genera Lactobacillus and Bifidobacterium. Other groups of the LAB (Streptococcus and Enterococcus), Bacillus, Propionibacterium and Saccharomyces are being used as probiotic microorganisms (Clinton et al., 2020).

The gastrointestinal tract (GIT) consists of diverse microorganisms mainly dominated by bacteria. Other archaeal domains, yeasts, and molds are also found in the GIT. Probiotic microorganisms are thought to have a wide range of health benefits (Sharma et al., 2021).

They exert antimicrobial activity against enteric pathogenic bacteria; stimulate mucosal and systemic immune responses of the host; provide anti colon cancer, anti-cholesterol, and anti-inflammatory effects; and improve nutritional status. They also reduce symptoms of diarrhoea, lactose into relance, and allergic reactions (Sharma et al., 2021).

So, multi strain or multispecies probiotic mixes are becoming more popular because they can give additive or even synergistic effects, as well as a broader spectrum of health advantages (Dias and Weimen, 2019).

The benefits of probiotic intervention in the gut microbiota in maintaining and restoring health are becoming better recognized, drawing increased scientific interest. The expected probiotic strains should be assessed for possession of physiological and genetic traits and technological properties beneficial to the host. Probiotic organisms must be able to live in the presence of gastric acid and bile, as well as adhere to and colonize the gastrointestinal epithelial layer. They also should be safe and non-pathogenic to the host (Urach, 2019).

#### 1.2 Statement of The Research Problem

This approach overcomes the problem of underestimation of the microbial diversity, which is often encountered when culture-dependent methods are used. Nevertheless, isolation and characterization

of lactic acid bacteria (LAB) inhabiting the cow milk and milk products are important to define the specific microbiota in order to maintain high standard of reproducibility of the final product. In fact, the characteristic presence of defined LAB in cow milk, together with that in milk products, is required for specific nutritional, organoleptic and physical properties of the derived cheese. Moreover, the search for LAB isolates with probiotic characteristic is still of great value due to scientific evidence, accumulated especially in the past decade, showing gut-derived effect of probiotic LAB strains on human intestinal health..

#### 1.3. Justification for the Research

The consumption of milk and fermented milk would reverse the adverse effect of the lower gut micro flora on the host animal. Milk products has a beneficial effect on the host's health. Milk products containing viable LAB have been used as prophylactics for treatment of intestinal infections in adults infected with Salmonella and lactose intolerance. Probiotics are live microorganisms that confer health benefits to the hosts by improving intestinal microbial balance. Some LAB species (Lactobacillus, Streptococcus, Enterococcus and Pediococcus) have been reported as active candidates for probiotic use in humans and animals by several researchers. Furthermore, the importance of the microbiota in the gut—brain axis is now largely explored. Therefore, on the light of the assessed value of some LAB strains, studies on isolates from cow milk and milk products may also be functional to the implementation of new promising probiotic strains.

# 1.4 Aims and Objectives

The aim of this study is to Isolate and identify Lactic Acid Bacteria from Cow Milk and Milk Products

The objectives of this research are to;

- i. Determine the bacterial load in the cow milk and milk products.
- ii. Isolate and identify lactic acid bacteria from cow milk and milk products.
- iii. Determine the sensory evaluation of LAB in the milk and milk products

#### **MATERIALS AND METHODS**

# 3.1 Study Area

Bodinga local government is located at latitude 12.8681° N and longitude 4.7456° E. It has an average elevation of 293 meters (433 feet). It is located along the Birnin kebbi road Sokoto National Highway. The temperature ranges from 25°C to 43°C depending on the season and the year. Red sandy loam, clayey soils, and alluvial soils are the major soil types existing. The climates is characterized by a hot summer, fair distributed rainfall during the season and generally dry weather during the rest of the year.

# 3.2 Study design

A combination of observational and experimental study designs were used in the isolation and identification of lactic acid bacteria in cow milk and milk products. This approach helps in gathering

comprehensive data on the presence, characteristics, and behavior of lactic acid bacteria in different contexts (Li T et al.,2020).

#### 3.3 Materials

The materials that were used for this practical are; culture medium, cow milk from Bodinga, milk products (hamdallah yogurt sold at Bodinga), conical flasks, test tubes, petri dish, stains racks, chemicals for gram staining reaction and biochemical tests, masking tapes, foil paper, cotton wool, flame burner or candle, wire loop, autoclave, hot air oven, incubator, distilled water weighing and colony counter.

# 3.4 Sample Collection and Sample Processing

This study was conducted at the Sokoto State University, Sokoto, Microbiological laboratory. Samples of cow milk and milk products (yogurt) were obtained from different local vendors, rearing farm within the Sokoto metropolis market located in Bodinga LGA, Sokoto state. A total of 7 cow milk and milk products were collected (2 raw cow milk samples from muhibat diary farm Bodinga) from two different cows, 2 milk samples from vending shop in Bodinga market, 1 sample of Hamdallah yogurt and 1 sample of milky plain yogurt from Babangida shop at Bodinga town and 1 milk sample from gidan manuga households. Early in the morning a total of 300ml to 500ml of milk and milk products samples were collected in clean and sterilize polythene bags or sterile bottles sealed labeled correctly and transferred to the microbiology laboratory at Sokoto State University Sokoto for microbiological analysis.

# 3.5 Nutrient Agar Medium preparation

The media was prepared according to the manufacturer's direction. This involves weighing fourteen grams (8.4g) of media for 15 plates, it was weighed using weighing balance and put into conical flask, (300ml) of water was added and the mixture was shaken, It was then heated to dissolve the powder and then sterilized by autoclaved at 121oc for 15minutes for sterilization (Cappuccino and Sherman, 2019).

# 3.5.1 Skim Milk Agar Medium preparation

The media was prepared according to the manufacturer's direction. These involves weighing of (8g) of the medium for 20 plates, it was weighed using weighing balance and put into conical flask, (400ml) of distilled water was added and it was mixed thoroughly, the mixture was then heated while stirring gently to dissolve the agar completely and boiling was avoid, after all components are dissolve then the medium was sterilized by autoclaving at 121oc for 15minutes. The sterilization media was allowed to cool at 45oc before pouring it into sterile Petri dishes (Adeniyi and Babalola, 2013).

#### 3.6 Serial Dilution

All the apparatus required for the practical ws sterilized before use. Serial dilution were carried out. Distilled water 9.0ml was transferred in to four sterile test tubes. One milliliter of the undiluted

sample was then collected with a sterile syringe and transferred into the first test tube containing distilled water. The mixtures was well shaken to get a homogenous mixture. One milliliter was pipetted from the first test tube and transferred into the second test tube containing distilled water. The process was repeated for subsequent dilutions, transferring 1 mL from the previous dilution tube to the next tube containing distilled water. Serial dilution procedure was carried out on each of the sample (Fawole and Oso, 2011).

# Microbiological Analysis

After the preparation of the media, it was allowed to solidify and ready for use. A small quantity, 0.1ml of the diluents from the last test tube was taken by the use of a sterile syringe, two to three drops of the diluents from the syringe was made on the surface of the solidified agar, and then spread on the surface of the agar using a sterilized bent glass rod and quickly and carefully covered. The same procedure were done on the rest of the sample; this procedure is known as spread plate method. Sub culturing was done to separate mixed colonies after incubation as described by (Tortora, Funke and Case, 2016).

# 3.7.1 Enumeration (Determination of bacteria load)

The total plate count for the growth of bacterial colonies on plates was observed by colony counter to count and record the number of colonies on each plate. The actual number of bacterial colonies was calculated as colony forming unit (CFU/ml) as described by (Horrigan and McCance, 2016).

# **Gram staining**

A smear was made by placing a drop of normal saline on a clean glass slide and sterilized wire loop was used to pick the culture which was mixed with the normal saline on the glass slide to make a smear. The smear was heat fixed by passing the slide over a flame. The glass slide was placed on the staining rack. The smear was covered with crystal violet to stained the bacterial cells and allowed to stand for one minute. It was washed with distilled water without blotting. This was followed by covering with iodine solution and allowed to stand for one minute, it was washed with distilled water. It was then flooded with acetone alcohol (decolorizer) and washed immediately to prevent excessive decolonization. Finally it was then flooded with saffranine and was allowed to dry. The dried slide was viewed under the microscope using oil lens (x100) by applying a drop of immersion oil. (Fawole and Oso, 2011).

#### 3.9 Biochemical Characterization of Isolates

# 3.9.1 Methyl Red Test

Methyl red test is employed to detect the production of sufficient acid during the fermentation of glucose. The test organism was incubated by inoculating the loopful of the organism into the methyl red medium that is glucose phosphate contained in a test tube and incubated at 37oc for 48 hours. After incubation, two drops of methyl red solution (methyl indicator) will be indicated by pink colour (Oyeleke and Manga, 2010).

# 3.9.2 Voges-Proskauer (VP) Test

Part of the same culture used for methyl red test was placed in another test tube before conducting methyl red test. Into this portion, 0.5 ml of Naphtol solution was added. 0.3 ml of 40% potassium hydroxide (KOH) aqueous solution was added and agitated. The test tube was slanted to solidify after which they will be examined (Oyeleke and Manga, 2010).

#### 3.9.3 Catalase test

This test demonstrates the presence of an enzyme catalase that catalyses the enzymatic breakdown of hydrogen peroxide to water with subsequent release of oxygen gas seen as bubbles. Test organism was inoculated into prepared nutrient agar and was incubated at 37oc for hours. After which 1ml 2-6% of hydrogen peroxide (H2O2) was added into the test tubes and the bubble was examined. Release of bubble indicates a positive reaction while no gas bubbles indicate a negative reaction (Oyeleke and Manga, 2008).

# 3.9.4 Coagulase Test

A slide or test tube with a small amount of sterile plasma was prepared. Using a sterile loop or needle, a small amount of the bacterial culture was transferred to the plasma. The bacteria and plasma was mix thoroughly, then the mixture was incubate at 37°C (body temperature) for a set period, usually around 4 hours, or until clotting is observed. (Oyeleke and Manga, 2008)

# 3.9.5 Starch Hydrolysis Test

The organism to be tested was inoculated on the agar and incubated for 24 hours. The plate was flooded with iodine and results was observed. Blue color indicates no hydrolysis while a clear zone indicates hydrolysis (Oyeleke and Manga, 2008)

#### 3.9.6 Citrate test

This test identified organisms capable of using sodium citrate as the sole carbon source and inorganic ammonium salts as the sole nitrogen source. The test is part of a series referred to as IMViC (indole, methyl red, Voges-Proskauer, and citrate), which is used to differentiate Enterobacteriaceae from other gram-negative rods. Koser citrate was prepared by weighing 2.5g of Sodium citrate, 1.5g of ammonium phosphate, 0.2g of Magnesium sulphate, 1g of potassium dehydrogenate phosphate and o.1g of bromothymol blue and dissolved in 1L of distilled water, homogenized and dispensed in test tubes then corked with cotton wool. A speck of each isolate was inoculated into Koser's citrate medium and incubated at 37°C for 72hours. A positive citrate was confirmed by formation of blue colour while the initial green colour denoted negative result (Oyeleke and Manga, 2010).

# **3.9.7 Motility**

A speck of each isolate was stabbed into triple sugar ion agar and incubated at 37°C for 24hours. Motility was observed by spreading the organism outwards from the stabbed area (Fawole and Oso, 2011).

#### 3.9.8 Urease test

Christensen urea agar was prepared by weighing 20g of plain agar, 1g of peptone, 1g of glucose, 0.1g of phenol red, 1.2g of disodium hydrogen orthophosphate and 5g of sodium chloride was dissolved in 100ml distilled water, heated to achieve total dissolution. The pH was adjusted to 6.8 using electrode pH meter to give yellow colour, dispensing into universal bottles and sterilized by autoclaving at 121°C for 15minutes. 5ml of 40%membrance sterile will be aseptically introduced into the universal bottle and then allowed to solidify in slanting position. A speck of each isolate was dipped into Christensen's urea agar and incubated at 37°C for 24hours. Liberation of red colour indicates urease positive while initial yellow colour indicates negative test (Oyeleke and Manga, 2013).

# 3.9.9 Triple Sugar Iron (TSI)

This medium contains three sugar namely glucose, sucrose and lactose. Some organism can ferment all three sugar present and produces acid which changes the colour of the indicator from red to yellow. The sugars and protein were attacked oxidatively to release ammonia. Through this media, the production of H2S was detected by the presence of a black colour in the media along the stabbed line. Motility was detected by the presence of growth along the area been stabbed by the straight wire loop. Gas production was detected by the presences of gas bubbles or crack on the agar in the test tube or complete disruption of the medium. Colonies from the sub-cultured plate was picked with a sterile straight wire loop and stabbed on the butt; streaked on the surface of the slope and was incubated at 370C for 48hrs (Chessbrough, 2010).

# 3.10 Analysis of sensory characterization of cow milk and milk product.

# 3.10.1 pH Measurement:

The pH meter and probe are clean and in good working condition. Then the pH meter was turned on and allow to warm up, the pH probe was rinsed with distilled water and gently blot dry with a lint-free tissue, the probe was immersed in the pH 7.0 calibration solution and the meter was adjusted to read 7.0, the process was repeated with pH 4.0 and 10.0 calibration solutions, the cow milk or milk product was homogenize to ensure consistency, the pH probe was immerse in the milk sample, ensuring the tip is fully submerged. Then the reading as allowed to stabilize, the pH value displayed on the meter will be recorded, these method is done by American Dairy Science Association

# 3.10.2 Sensory evaluation:

The samples was homogenize to ensure uniformity, then the samples are labelled to avoid bias, the sample was presented in sterile containers, then each sample was served in sufficient quantity for multiple evaluation, the sample is evaluated base on specific sensory attributes which are appearance, aroma, taste, texture, and acceptability (Meilgaard et al., 2015)

**Appearance:** To look for signs of curdling, separation, or unusual color changes.

**Aroma:** To detect sour, acidic, or off-door's that are indicative of fermentation.

Taste: Identify any sourness, bitterness, or off-flavors that differ from the fresh product.

**Texture:** To check if there is any change in consistency, such as increased thickness, graininess, or smoothness.

#### **CHAPTER FOUR**

#### 4.0 RESULTS

The results of the research on the isolation and identification of lactic acid bacteria from cow milk and milk product (cheese), are represented in Tables 4.1 to 4.5.

Table 4.1 Presents the dilution factor of the bacterial colony counts on the cow milk and milk products which include the diluting factor 10<sup>2</sup>, number of inoculum, and size of the cfu/ml.

Table 4.2 Present the cultural characteristics of the bacterial isolated from cow milk and milk product which includes the color, shape, size, elevation, and texture of the colonies and then the gram staining reaction and arrangement of the bacteria.

Table 4.3 Presents the percentage of frequency occurrence of the bacterial isolates from the cow milk and the milk products Which includes Lactobacillus acidophilus and Lactococcus lactis which had the highest percentage of 28.51%, each while Lactobacillus bulgaris, Lactobacillus casei and Lactobacillus planetarium had average of 14.28% each.

Table 4.4 Shows the biochemical characterization of bacterial isolates. The identified bacterial isolates include, Lactobacillus acidophilus, Lactobacillus bulgaris, Lactobacillus casei, Lactobacillus planetarium and Lactococcus lactis

Table 4.5 Presents the sensory characteristic of the cow milk and milk product (yogurt), by using sensory evaluation to detect the; appearance, aroma, taste, texture, and mouth feel of the cow milk and milk product.

Table 4.1: The Dilution factor and Total Bacterial Colony Count From Cow Milk And Milk Products.

<b>Isolate Code</b>	Dilution	Number of Colonies	Inoculums (ML)	Size Cfu/ml
A	$10^{2}$	27×4= 108	0.5	$108 \times 10^2$
В	$10^{2}$	22×4= 88	0.5	$88 \times 10^{2}$
C	$10^{2}$	39×4= 156	0.5	$156 \times 10^2$
D	$10^{2}$	44×4=176	0.5	$176 \times 10^2$
E	$10^{2}$	17×4= 68	0.5	$68 \times 10^2$
F	$10^{2}$	52×4= 208	0.5	$208 \times 10^{2}$
G	$10^{2}$	12×4= 48	0.5	48×10 <sup>2</sup>

**KEY:** A = Milk product (Plain yogurt), B = Milk product (Hamdala Yogurt), C = Cow Milk from (Bodinga Market), D = Cow Milk From Vendor Shop at Bodinga, E = Cow Milk From Muhibat Farm at Bodinga, F = Cow Milk From Bodinga Market 2, G = Cow Milk From Vendor Shop Bodinga 2.

S/N	Color	shape	Size	Elevation	Texture	Arrangement
A.	Whitish	round	1.2mm	flat	dry	chain
B.	Whitish	round	1.2mm	flat	creamy	chain
C.	Yellowish	round	1.7mm	flat	mucoid	chain
D.	Whitish	round	2.0mm	flat	creamy	chain
E.	Whitish	round	1.4mm	flat	creamy	chain
F.	Whitish	round	1.3mm	flat	creamy	chain
G.	Whitish	round	1.8mm	flat	creamy	chain

**Table 4.2 The Cultural Characteristics of the Bacterial Isolates** 

**KEY:** + = Positive - = Negative A = Milk product (Plain yogurt), B = Milk product (Hamdala Yogurt), C = Cow Milk from (Bodinga Market), D = Cow Milk from Vendor Shop at Bodinga, E = Cow Milk From Muhibat Farm at Bodinga, F = Cow Milk From Bodinga Market 2, G = Cow Milk From Vendor Shop Bodinga 2.

Table 4.3 Shows The Mophological and Biochemical Characterization Of Bacterial Isolates
From Cow Milk And Milk Products (yogurt)

S/N	Gram	Cat	Coa	Glu	Lac	Suc	H <sub>2</sub> S	Gas	Cit	Ure	Mr	Vp	Ind	Mot	Sta	Suspected organism
1.	+ rod	+	+	+	+	+	+	+	+	+	+	-	-	-	-	Lactobacillus acidophilus
2.	+ rod	+	-	+	+	+	-	+	+	+	+	-	-	-	-	Lactobacillus bulgaris
3.	+ rod	+	-	+	+	-	-	+	+	+	-	+	-	-	+	Lactobacillus casei
4.	+ rod	+	+	+	+	+	+	+	+	+	+	-	-	-	+	Lactobacillus planetarium
5.	+cocci	+	-	+	+	-	-	+	+	+	-	+	-	-	-	Lactococcus lactis
6.	+ rod	+	-	+	+	+	-	+	+	+	+	-	-	-	-	Lactobacillus acidophillus
7.	+cocci	+	-	+	+	-	-	+	+	+	-	+	-	-	-	Lactococcus lactis

**KEY**= += Positive, - = Negative Gram Reaction = Gram Staining Reaction, GLU = Glucose, LAC = Lactose, SUC = Sucrose, CAT = Catalase, CIT = Citrate, MOT = Motility, IND= Indole, URE= Ureases, COA = COAGULASE, H2S = Hydrosulphide, GAS = Gas Production, STA = Starch Hydrolysis Test.

Table 4.4; Frequency and Percentage of Occurrence of the Bacteria Isolated from the Samples.

S/N	Identified Organism	Frequency of Occurrence	Percentage of Occurrence
1.	Lactobacillus acidophiles	2	28.51%
2.	Lactococcus lactis	2	28.51%
3.	Lactobacillus bulgaris	1	14.28%
4.	Lactobacillus casei	1	14.28%
5.	Lactococcus lactis	1	14.28%
Total		7	100%

Table 4.5 Presents The Sensory Characteristic of The Cow Milk And Milk Product (Yogurt), pH Measurement and Sensory Evaluation

Sample	Appearance	Aroma	Taste	Texture	Mouth feel	pН
<b>A.</b>	White and clear	Sweetness	Sweet	Thick	Thick	4.6
В.	White and clear	Sweetness	Slightly sour	Thick	Slightly watery	4.6
<b>C.</b>	White but blur	Sweet	Slightly sour	Smooth and creamy	Watery	4.8
D.	Whitish	Sweetness	Sour	Watery	Watery	4.0
<b>E.</b>	White and clear	Sweetness	Slightly sour	Slightly thick	Watery	5.3
F.	White and clear	Sweetness	Sweet	Thick	Thick	3.9
G.	Creamy white	Creamy	Sweet	Smooth	Refreshing with flavour	4.2

**KEY:** A = Milk product (Plain yogurt), B = Milk product (Hamdala Yogurt), C = Cow Milk from (Bodinga Market), D = Cow Milk From Vendor Shop at Bodinga, E = Cow Milk From Muhibat Farm at Bodinga, F = Cow Milk From Bodinga Market 2, G = Cow Milk From Vendor Shop Bodinga 2.

# **CHAPTER FIVE**

#### 5.0. DISCUSSION CONCLUSION AND RECOMMENDATION

# **5.1 DISCUSSION**

Lactic acid bacteria (LAB) are currently the subject of extensive international research due to their critical involvement in most fermented foods, as well as their potential to create antimicrobial chemicals that promote probiotic characteristics including antitumor activity, alleviation of lactose intolerance, a decrease of serum cholesterol, gut microflora stabilization, and stimulation of the

immune system. In the production of fermented milk, the LAB strain produced for exopolysaccharides is used to improve their viscosity and smoothness. Some LAB strains are known to produce the presumed effects of mannitol which promote health. Probiotics are live bacteria or yeasts that, when given in sufficient proportions, provide a health benefit to the host.

This study sought to isolate and identify LAB from cow milk and milk product. A total of seven samples were used for this study. The identification of the LAB was performed through morphological characteristics and biochemical tests and the entire LAB was identified to its generic level. Firstly the enumeration of the LAB from different cow milk and milk products was revealed that Lactobacillus spp were presumptively enumerated as  $1.08 \times 104$ ,  $8.8 \times 103$ ,  $3.9 \times 103$ ,  $4.4 \times 103$  and  $2.08 \times 104$  cfu/ml from the raw cow milk, and yogurt, while load of Lactococcus spp was counted as  $6.8 \times 103$  and  $4.8 \times 103$  cfu/ml from raw cow's milk, and yogurt, respectively. These findings were in comparison with the earlier report of Khedid et al., (2014) from Morocco, who reported that Lactobacillus spp counted load were  $7.5 \times 104$  cfu/ml from cow milk. Moreover the Lactococcus spp counted load in this present study was higher than the report of Khedid et al., (2014), which is  $7.6 \times 104$  cfu/ml but the both results are within the standard which is 105 cfu/ml based on IDF (2018).

The morphological and cultural characteristics of the bacterial colonies in this study revealed are whitish in color, small to large size, and round on nutrient agar media and also, the gram staining characteristics of the bacteria revealed that all of the bacteria were Gram-positive, while some were rod and some were spherical shaped, and have different arrangements as follows: Lactococcus spp appeared spherical and in pairs and some in chain arrangement, while Lactobacillus spp appeared more in rod and in pairs.

From the result of biochemical characteristics (Table 4.3), a total of 5 bacteria were identified and categorized into two different species of LAB based on their percentage of occurrence (Table 4.4) include; (28.51%) of the bacteria identified were Lactobacillus acidophilus and Lactococcus lactis with the same percentage while lactobacillus planetarium, lactobacillus bulgaricus, and lactobacillus casei were found in average percentage of (14.8%). However, in yogurt Lactobacillus spp were found in a higher proportion than Lactococcus spp while in the case of cow milk, Lactococcus spp were found in average proportion. This is in accordance with the findings by vantsawa et al., (2017) carried out at unguwar rimi area kaduna who identified a higher proportion of Lactobacillus spp such as Lactobacillus acidophilus, Lactobacillus bulgaricus, lactobacillus salivarius and lactobacillus rhamnosus from fermented cow milk (nono).

Several research works have examined the findings of this study e.g the research conducted by Savadogo et al., (2021) and Harun-ur-Rashid et al., (2021) where they were able to identify six genera of LAB from traditional fermented milk, including Leuconostoc, Lactococcus, Lactobacillus, Enterococcus, Streptococcus, and Pedicoccus spp. Similarly Abdel Gawad et al., (2019) have also identified five different genera of LAB: Lactobacillus at (30%), Leuconostoc at (26%), Enterococci at (20%), Streptococcus at (18%), and Aerococcus spp at (18%) in cow milk. Also, a comparable figure was reported by Abdullah and Osman, (2019) where the genus Lactobacillus was dominant in all samples tested (69.23%) followed by the genus Lactococcus (19.23%) and the genus Pedi coccus (11.53%).

The sensory characteristic and pH measurement of the cow milk and milk product (Table 4.5) was examined and the sensory evaluation was obtained by the physical appearance, aroma, taste, texture and mouth feel of the cow milk and milk product, while the pH level of the all the cow milk and milk products ranges from 3.9 to 5.3 which is between the standard pH level of cow milk and milk products for consumption. IDF., (2018) standard. Moreover, this study has similar findings with El-Shafei et al. (2018) and Abd El Gawad et al., (2019) who identified a higher proportion of Lactobacillus spp from milk products, Lactobacillus spp can survive in a highly acidic environment with a pH of 4 to 5 or even lower, and it is Lactobacillus that is responsible for the final stages of fermentation in the goods as a result of these traits. This also demonstrated that Lactobacillus spp survive in low pH environments.

El-Shafei et al. (2018) also reported the preponderance of Lactobacillus spp was higher among isolated LAB because milk and milk products constitute a varied mixture of diverse microorganisms. Furthermore, cow milk, which is widely used to make fermented milk, may promote the growth of these species. Lactobacillus spp can survive in a highly acidic environment with a pH of 4 to 5 or even lower and it is Lactobacillus spp that is responsible for the final stages of fermentation in products because of their qualities. This also demonstrated that Lactobacillus spp survive in low pH environments

#### **5.2 CONCLUSION**

In conclusion, lactic acid bacteria were successfully isolated from cow milk and milk product. The characterization of isolates on the basis of microscopic analysis and biochemical properties (phenotypic characterization) is very useful being the most widely recognized and accepted method.

Lactic acid bacteria (LAB) have long been consumed by people in several fermented foods such as dairy products. A study was conducted on lactating dairy cows to isolate and characterize LAB from dairy products found in and around Sokoto metropolis cow milk and milk products were randomly collected. The current study revealed that most of the isolated LAB species were grouped into two genera including Lactobacillus and Lactococcus spp, and the Lactobacillus were the most common LAB isolated from various cow milk and milk products among the identified genera. Moreover, Lactococcus spp were the commonly isolated from cow milk only, while Lactobacillus spp were found in both in cow milk and milk products (yogurt). Based on the enumeration of LAB, Lactobacillus and Lactococcus spp were counted in higher proportion from yogurt and cow milk.

#### **5.3 RECOMMENDATION**

I therefore recommend that further study on the identification and characterization of LAB to strain level should be done. The health and economic benefits of the identified LAB should be given more attention in order to improve community health. The possible application of these LAB as probiotics should be further tested on different species of experimental animals.

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