ANTIBIOGRAM OF BACTERIA ISOLATED IN RAW MILK SOLD IN MAKURDI METROPOLIS, BENUE STATE, NIGERIA

ABSTRACT

The antibiogram profile of bacteria isolated from unpasteurized milk source from selected markets in the Makurdi metropolis was analyzed. A total of 300 samples of unpasteurized milk were examined. Out of which 222(74%) samples tested positive for various bacteria isolates. The most frequently identified bacterium was *Escherichia coli*, which constituted 72(32.4%) followed by *Staphylococcus aureus* 42(18.92%), *Salmonella species* were the third most common, representing 30(13.51%), *Pseudomonas aeruginosa* and *Proteus species*both had 16(7.21%), *Bacillus species* 20(9.91%), and *Streptococcus species* 20(9.01%). The antibiogram results revealed that all isolated bacteria showed significant sensitivity to Chloramphenicol and Gentamicin. These findings highlight the need for government support for local milk producers to obtain resources and establish collection centers with adequate pasteurization facilities, thereby reducing the risks linked to the consumption of contaminated milk products. It is crucial that milk is pasteurized immediately after collection to lower bacterial loads, especially pathogenic strains. Further research is essential to devise effective strategies to tackle the issues related to unpasteurized milk.

Keywords: Bacteria, Antibiotics, Raw milk,liquid food

1. INTRODUCTION

Raw milk is defined as milk that has not been subjected to pasteurization, a process that involves heating liquid food to eliminate harmful microorganisms, ensuring it is safe for consumption (Beecher and Cookson, 2016). Proponents of raw milk contend that it provides benefits such as superior flavor, enhanced nutritional content, and bolstered immune support. However, the medical community has expressed concerns about its safety, particularly regarding the potential for infections (Smith *et al.,* 2012). The regulation and availability of raw or unpasteurized milk vary widely across different regions worldwide. In the United States, certain dairy producers have adopted low-temperature pasteurization methods, asserting that these techniques produce a product similar to raw milk (Smith *et al.,* 2012).

Raw milk is recognized as a highly nutritious food, specifically designed to support the growth of young individuals and to enhance the dietary balance of adults. It boasts a higher concentration of essential nutrients compared to many other individual food sources (Oliver *et al.,* 2005). Throughout history, milk and dairy products have been celebrated as "the most nearly perfect food" due to their impressive nutritional composition. Milk is an excellent source of calcium and phosphorus and is abundant in vitamins A, B1, B6, and B12, all of which are crucial for the development of bones and teeth (Oliver *et al.,* 2005). Nevertheless, both raw and processed milk can create a favorable environment for the growth of various microorganisms, which may result in spoilage or pose risks of infections and intoxications for consumers (Oliver *et al.,* 2005).

The existence of contaminating microorganisms, especially pathogenic bacteria, in milk presents serious public health issues. Poor hygiene practices among individuals handling raw milk can lead to the introduction of dangerous pathogens into the product. Given that raw milk is not processed further before it is consumed, it can pose health risks to consumers (Adeyemi and Umar, 1994). The rise of antibiotic-resistant pathogenic bacteria intensifies public health challenges globally, constituting a growing threat to health on an international scale (Levy, 2001).

2. MATERIALS AND METHODS

2.1 Study Area

The study was carried out in Makurdi, the capital city of Benue State,Nigeria. This city is situated between latitudes 7° 47' and 10° 00' North, and longitudes 6° 25' and 8° 81' East of the equator. It is bordered to the north by Guma Local Government Area, to the south by Gwer East

Local Government, to the southwest by Gwer-West Local Government Area, and to the northwest by Doma Local Government Area in Nasarawa State. Makurdi lies within the Benue Valley along the banks of the Benue River. The town is a vital hub on the North-South transportation corridor, with access via both road and rail, connecting Nasarawa and Enugu States, and encompasses a total land area of approximately 810 square kilometers (National Population Commission, 2009; Mngutyo and Ogwuche, 2013; Olayinka *et al.*, 2013**).**

2.2 Study Population

The research concentrated on local milk sellers, specifically Fulani women, who market cow milk in Makurdi. Unpasteurized milk samples were gathered from multiple vendors throughout the Makurdi area, ensuring that at least two samples of raw cow milk were collected from each seller.

2.3 Sample Collection

A total of 300 raw milk samples were collected from various sites in Makurdi,specifically North Bank Market, Wurukum Market, Wadata Market, High Level Market, Kanshio Market, and Fiidi Market. Approximately 100 mL of each sample was aseptically placed into sterile containers, utilizing a sample collector ice box kept at 4°C. Each sample was distinctly labeled for identification purposes and was swiftly transported to the Research Laboratory of Medical Microbiology and Parasitology at Benue State University Teaching Hospital.

2.4 Determination of Total Microbial Counts in Milk

The total colony counts of microorganisms in the raw milk samples were assessed through the standard plate count method (Sanders, 2012). A series of dilutions was conducted on samples collected from different locations, and the diluted samples were then plated on Plate Agar utilizing the pour plate technique. The plates were incubated at 37°C for a duration of 24 hours to evaluate the average microbial loads present in each sample.

2.4 Isolation and identification of bacteria isolates

Standard bacteriological techniques were utilized for bacterial isolation, following the guidelines set forth by Cheesbrough (2006). Samples were serially diluted and subsequently inoculated using the pour plate method on MacConkey Agar, Chocolate Agar, Eosin Methylene Blue (EMB) Agar, Mannitol Salt Agar (MSA), and Salmonella-Shigella Agar (SSA) to initiate the preliminary identification of the isolates. MacConkey Agar facilitated the isolation of lactose-fermenting gram-negative bacteria, while Chocolate Agar was employed for the cultivation of fastidious organisms. Eosin Methylene Blue Agar was specifically used for the selective isolation of enteric coliforms, Mannitol Salt Agar targeted salt-tolerant bacteria, and Salmonella-Shigella Agar was designated for the isolation of enteric bacilli. All plates were incubated at 37°C for 24 hours, with identification based on cultural, morphological, and biochemical characteristics as outlined by Holt (1994).

2.6 **Antibiotic Susceptibility Testing**

Antibiotic susceptibility testing was conducted using the Kirby-Bauer disc diffusion method. This testing aimed to assess the antibiotic resistance and susceptibility profiles of bacterial isolates (Kirby *et al.,* 1966). Bacteria were taken from agar slants and inoculated into tryptose soy broth, followed by incubation at 37°C. Fresh overnight cultures were utilized for the antibiotic sensitivity assessments. An aliquot of 0.5 mL from each isolate suspension was evenly spread on Mueller Hinton agar (Oxoid, England) using a sterilized swab. Antibiotic discs were then carefully placed on the inoculated agar to ensure proper contact with the surface, and the plates were incubated aerobically at 37°C for a duration of 18 to 24 hours (CLSI, 2019). The antimicrobial susceptibility test involved nine different antibiotics: erythromycin (15 µg), penicillin (10µg), gentamicin (10 µg), trimethoprim-sulfamethoxazole (25 µg), chloramphenicol (30 µg), vancomycin (30 µg), tetracycline (30 µg), cefoxitin (30 µg), and ciprofloxacin (5 µg). The susceptibility of the bacterial isolates was determined by measuring the inhibition zone diameters for each antibiotic, with results classified as susceptible, intermediate, or resistant according to the interpretive criteria established by the Clinical and Laboratory Standards Institute (CLSI, 2019).Statistical Analyses

2.7 Statistical Analysis

The data collected were examined utilizing the Statistical Package for Social Sciences (SPSS) version 20.0 software (SPSS, 2012). The analysis included the calculation of averages, proportions (percentages), two-way analysis of variance, and the Least Significant Difference test to differentiate between means.

3. RESULTS

The isolates were confirmed using different biochemical test as shown in table 1. The occurrences of bacterial isolates in unpasteurized milk gotten from samples collected from different markets in Makurdi metropolis showed that, out of 300 samples screened, 222 (74%) were positive for bacterial isolates. Wadata MKT had the highest percentage of bacterial isolates

44 (88%), followed by North Bank MKT 42 (84%), High Level MKT 38 (76%), Wurukum MKT 36 (72%), Fiidi MKT 32 (64%), while Kanshio MKT had the least percentage 30 (60%) as shown in table 2.

Table 3 shows different prevalence of various bacterial isolates in unpasteurized milk gotten from different markets in Makurdi metropolis. *Escharichia coli* in North bank MKT had highest percentage of (42.9%), followed by Wadata MKT (36.4%), while Fiidi MKT had the least prevalence of (25.0%). *Staphylococcus aureus* in North Bank MKT (23.8%) was highest in prevalence, followed by Wadata MKT (22.7%) while Fiidi MKT had the least prevalence of (12.5%) of bacterial isolates.*Salmonellaspecies* in Fiidi MKT was highest with prevalence of (25.0%), followed by Kanshio MKT (20.0%), while North bank MKT had the least prevalence of (9.5%). *Pseudomonas aeruginosa* had the highest prevalence of (11.11%) in Wurukum MKT, followed by (10.5%) in High Level MKT while Wadata MKT had the least prevalence of (4.5%).*Bacillus cereus* was found high in Kanshio MKT (13.33%), followed by Wurukum MKT (11.11%) while Fidi MKT had the least prevalence of (6.25%). *Proteus species* was found high in Fidi MKT (12.5%), followed by High Level MKT (10.5%) while Wadata MKT had the least prevalence percentage of 4.5%. *Streptococcus species* was found high in Fidi MKT (12.5%), followed by Wurukum MKT (11.11%) while North bank MKT had the least prevalence of (4.8%).

Out of 222 bacterial isolates, antibiotic susceptibility test were performed on 72(32.4%) isolates of *E.coli.Escherichia coli* isolates were found to be highly susceptible to Ciprofloxacin (100%), followed by Gentamycin (86.1%). However, resistance to Penicillin (96.1%), followed by Cephoxitin (58.33%) shown in table 4. *Staphylococcus aureus*were found to be highly susceptible to Ciprofloxacin (100%), followed by Gentamycin, erythromycin and vancomycin

(83.33%). However, Resistance to Cephoxitin (88.09%), followed by Penicillin (71.4%) shown in table 5. *Salmonella Spp* isolates were found to be highly susceptible to Ciprofloxacin (100%), followed by Gentamycin (86.7%). However, resistance to Tetracycline(86.7%),followed by Vancomycin (83.3%) as shown in table 6.*Pseudomonas aeruginosa* isolates were found to be highly susceptible to Ciprofloxacin (100%), followed by Chloramphenicol (87.5%). However, resistance to Vancomycin and Penicillin (100%), followed by followed by Cephoxitin (68.7%) shown in table 7. *Bacillus cereus* isolates were found to be highly susceptible to Ciprofloxacin, Trimethoprim-sulfamethoxazole and Gentamycin (90.91%), followed by Chloramphenicol (86.4%) while Erythromycin (81.82%) was the fourth antibiotics that are susceptible to *Bacillus Spp* isolates. However, resistance to Cephoxitin (90.91%), followed by Penicillin (86.4%) and Vancomycin (81.82%) merged the third antibiotics that were resistance to *Bacillus cereus* shown in table 8.*Proteus Spp* isolates were found to be highly susceptible to Ciprofloxacin (100%), followed by Chloramphenicol (87.5%) while Gentamycin and Trimethoprim-sulfamethoxazole (75.0%) came third. However, resistance to Vancomycin (75.0%), Cephoxitin (75.0%), Tetracycline (75.0%) and Penicillin (75.0%) shown in table 9.*Streptococcus Spp* isolates were found to be highly susceptible to Ciprofloxacin and Gentamycin, Chloramphenicol (90.0%) followed by Trimethoprim-sulfamethoxazole (80.0%) However, resistance to Penicillin, Cephoxitin and Vancomycin (90.0%), followed by Tetracycline (85.0%) is shown in table 10.

Table 1: Biochemical tests for the identifications of isolates obtained from Unpasteurized Milk in Makurdi Metropolis.

	Isolates

Biochemical Tests	*Escherichia Coli*	*Staphylococcus aureus*	*Salmonella typhi*	*Pseudomunas aeruginosa*	*Bacillus cereus*	*Proteus spp*	*Streptococcus pyogenes*
Catalase test	+	+	+	+	+	+	−
Coagulasetest	−	+	−	−	−	−	−
Citrate utilization test	−	+	−	+	+	+	−
MR test	+	+	+	−	−	+	−
VP test	−	+	−	−	+	−	
Indole test	+	−	−	−	−	−	−
Oxidase test	−	−	−	+	−	−	−
TSI A test Glu	+	+	+	−	+	+	+
Lactose	+	+	−	−	−	−	+
Urease test	−	+	−	−	−	+	−

Key: − Negative, + Positive

Table 2: Occurrence Frequency of Bacterial Isolates from Unpasteurized Milk purchased from Selected Markets in Makurdi metropolis in percentage (%).

Markets	Sample size	No. of sample positive for bacteria isolation in unpasteurized milk
North Bank MKT	50	42 (84%)
High Level MKT	50	38 (76%)
Wadata MKT	50	44 (88%)
Kanshio MKT	50	30 (60%)
Fidi MKT	50	32 (64%)

Wurukum MKT	50	36 (72%)

Key: MKT- Market

Table 3: Prevalence of different bacterial isolated from unpasteurized Milk samples with respect to Markets in Makurdi metropolis in percentage.

				Bacterial	Pathogens			
Markets	No. of isolates	*E.coli*	*S. aureus*	*Salmonella spp*	*p. aeruginosa*	*Bacillus cereus*	*Proteus spp*	*Sreptoccus spp*
NBMKT	42	18(42.9%)	10(23.8%)	4(9.5%)	2(4.8%)	4(9.5%)	2(4.8%)	2(4.8%)
HLMKT	38	10(26.3%)	8(21.1%)	4(10.5%)	4(10.5%)	4(10.5%)	4(10.5%)	4(10.5%)
WAMKT	44	16(36.4%)	10(22.7%)	6(13.6%)	2(4.5%)	4(9.1%)	2(4.5%)	4(9.1%)
KMKT	30	10(33.3%)	4(13.3%)	6(20.0%)	2(6.7%)	4(13.33%)	2(6.7%)	2(6.7%)

FMKT	32	8(25.0%)	4(12.5%)	8(25.0%)	2(6.25%)	2(6.25%)	4(12.5%)	4(12.5%)
WUMKT	36	10(27.8%)	6(16.7%)	6(16.7%)	4(11.11%)	4(11.11%)	2(5.6%)	4(11.11%)

Key: NBMKT- North bank Market, HLMKT- High Level Market, WAMKT- Wadata Market, KMKT – Kanshio Market, FMKT- Fidi Market, WUMKT- Wurukum Market and *spp*- species.

Table 4: Antibiotic Sensitivity Pattern of *Escherichia coli* (n=72) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis in percentage (%).

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	60(83.33%)	6(8.33%)	6(8.33%)
Penicillin	10(13.9%)	-	62(86.1%)
Gentamycin	62(86.1%)	-	10(13.9%)
Trimethoprim/sulfamethoxazole	55(76.4%)	5(6.9%)	12(16.7%)

Chloramphenicol	50(69.44%)	10(13.9%)	12(16.7%)
Vancomycin	60(83.33%)	-	12(16.7%)
Tetracycline	55(76.4%)	2(2.8%)	15(20.8%)
Cephoxitin	30(41.7%)	-	42(58.33%)
Ciprofloxacin	72(100%)	0(0%)	0(0%)

Table 5: Antibiotic Sensitivity pattern of *Staphylococcus aureus* (n=42) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis in percentage (%).

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	35(83.33%)	2(4.8%)	5(11.9%)
Penicillin	6(14.3%)	6(14.3%)	30(71.4%)
Gentamycin	35(83.33%)	-	7(16.7%)

Trimethoprim/sulfamethoxazole	27(6436%)	5(11.9%)	10(23.8%)
Chloramphenicol	30(71.4%)	4(9.5%)	8(19.05%)
Vancomycin	35(83.33%)	-	7(16.7%)
Tetracycline	30(71.4%)	(7.14%)	9(21.43%)
Cephoxitin	5(11.91%)	-	37(88.09%)
Ciprofloxacin	42(100%)	0(0%)	0(0%)

Table 6: Antibiotic Sensitivity Pattern *of Salmonella Spp* (n=30) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis.

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	20(66.9%)	2(6.7%)	8(26.7%)
Penicillin	7(23.33%)	-	23(76.7%)
Gentamycin	26(86.7%)	-	4(13.3%)
Trimethoprim/sulfamethoxazole	20(66.7%)	5(16.7%)	5(16.7%)

Chloramphenicol	25(83.3%)	-	5(16.7%)
Vancomycin	5(16.7%)	-	25(83.3%)
Tetracycline	4(13.3%)	-	26(86.7%)
Cephoxitin	6(20.0%)	3(10.0%)	21(70.0%)
Ciprofloxacin	30(100%)	0(0%)	0(0%)

Table 7: Antibiotic Sensitivity Pattern of *Pseudomonas aeruginosa* (n=16) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis in percentage (%).

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	12(75.0%)	4(25.0%)	0(0%)
Penicillin	0(0%)	-	16(100%)
Gentamycin	10(62.5%)	-	6(37.5%)
Trimethoprim/sulfamethoxazole	7(43.75%)	4(25.0%)	5(31.25%)

Chloramphenicol	14(87.5%)	-	2(12.5%)
Vancomycin	0(0%)	-	16(100%)
Tetracycline	7(43.75%)	4(25.0%)	5(23.25%)
Cephoxitin	5(31.25%)	-	11(68.75%)
Ciprofloxacin	16(100%)	0(0%)	0(0%)

Table 8: Antibiotic Sensitivity Pattern *Bacillus cereus* (n=22) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis.

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	18(81.82%)	2(9.09%)	2(9.09%)
Penicillin	3(13.6%)	-	19(86.4%)
Gentamycin	20(90.90%)	-	2(9.09%)
Trimethoprim/sulfamethoxazole	20(90.90%)	1(4.5%)	1(4.5%)
Chloramphenicol	19(86.4%)	-	3(13.6%)

Vancomycin	2(9.09%)	2(9.09%)	18(81.82%)
Tetracycline	4(18.18%)	1(4.5%)	17(77.3%)
Cephoxitin	2(9.09%)	-	20(90.91%)
Ciprofloxacin	20(90.91%)	-	2(9.09%)

Table 9: Antibiotic Sensitivity Pattern *of Proteus Spp* (n=16) from Unpasteurized Milk obtained from Selected Markets Makurdi metropolis.

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	11(68.75%)	2(12.5%)	3(18.75%)
Penicillin	4(25.0%)	-	12(75.0%)
Gentamycin	12(75.0%)	-	4(25.0%)
Trimethoprim/sulfamethoxazole	12(75.0%)	2(12.5%)	2(12.5%)
Chloramphenicol	14(87.5%)	-	2(12.5%)
Vancomycin	4(25.0%)	-	12(75.0%)

Tetracycline	2(12.5%)	2(12.5%)	12(75.0%)
Cephoxitin	4(25.0%)	-	12(75.0%)
Ciprofloxacin	16(100%)	0(0%)	0(0%)

Table 10: Antibiotic Sensitivity Pattern *of Streptococcus Spp* (n=20) from Unpasteurized Milk obtained from Selected Markets in Makurdi metropolis.

Antimicrobial agents	Susceptible	Intermediate	Resistant
Erythromycin	14(70.0%)	4(20.0%)	2(10.0%)
Penicillin	2(10.0%)	-	18(90.0%)
Gentamycin	18(90.0%)	-	2(10.0%)
Trimethoprim/sulfamethoxazole	16(80.0%)	2(10.0%)	2(10.0%)
Chloramphenicol	18(90.0%)	-	2(10.0%)
Vancomycin	2(10.0%)	-	18(90.0%)
Tetracycline	2(10.0%)	1(5.0%)	17(85.0%)

Cephoxitin	2(10.0%)	-	18(90.0%)
Ciprofloxacin	18(90.0%)	-	2(10.0%)

4. DISCUSSION

Pathogenic bacteria represent a considerable risk to global public health. The nutrient-dense nature of milk supports the growth of a variety of microorganisms, including harmful strains (Saeed *et al.,* 2009). Pareke and Subhash (2008) indicate that factors such as the health of the animals, the cleanliness of milking equipment, and environmental conditions play a role in the contamination of fresh raw milk.

The results of this study indicated a significant degree of bacterial contamination in the analyzed samples. This finding is consistent with the work of Syed et al. (2014), who noted similarly high contamination levels in unpasteurized milk sourced from Beed City. The contamination levels found in this study align with those reported by Makwin*et al.* (2014) in Keffi, although they differ from the results obtained by Laba and Udonsek (2013) in Ilorin.

This research underscores the identification of bacteria pathogens in raw milk source from Makurdi city. The highest prevalence of *Escherichia coli* was observed in North Bank market, with a detection rate of (42.9%), followed by Wadata market (36.4%) while Fiidi market had the least (25.0%) as the most prevalent bacteria similar to the finding of Jyoti *et al.* (2014) and Kewler*et al*. (1992) and contrary to that of Makwin*et al*. (2014) in Keffi and higher than that of Olatunji *et al.* (2013). The isolation of *Staphylococcus aureus* was higher in North bank market (23.8%), followed by wadata market (22.7%) and Fiidi market had the least (12.5%) in this study which agrees with that of Makwin*et al*. (2014), Olatunji *et al.* (2013), Jyoti *et al.* (2014) and Anklo and Sterneojo (2006) who isolated 30%, 26.7%, 33.33%, 20.4% and 15% respectively from unpasteurized milk. The presence of Salmonella species indicates the potential for typhoid, posing a significant health risk to consumers. The prevalence of *Salmonella species* (25.0%) in Fiidi market was closely followed in Kanshio market (20.0%) and North bank market had the least (9.5%) which is in agreement with the findings of Jyoti *et al.* (2014) and Makwin*et al.* (2014). The lower percentages were obtained among *P. aerugunosa* (11.11%), *Bacillus spp* (13.33%), *Proteus spp* (12.50%) and *Streptococcus spp* (12.5%) which agreed with Olatunji *et al.* (2013), Donkor *et al.* (2007), Laba and Udonsek (2013) and Makwin*et al.* (2014). On analyzing the data, the isolates were found to be statistically non-significant ($p>0.5$).

The study identified 72 isolates of *Escherichia coli* that displayed notable susceptibility to various antibiotics, showing complete sensitivity to Ciprofloxacin (100%), followed by Gentamycin at 86.1%, and both Vancomycin and Erythromycin at 83.33%. Tetracycline and

Trimethoprim-sulfamethoxazole had a susceptibility rate of 76.4%. In contrast, resistance was observed against Penicillin (86.1%) and Cephoxitin (58.33%). The significant resistance to penicillin aligns with findings from Sileshi and Munees (2016) in Ethiopia and Martha *et al.* (2016) in Tanzania, as well as observations by Idriss *et al*. (2014) in Slovakia and Belayneh et al. (2014) in Ethiopia. The high sensitivity to Ciprofloxacin, Gentamycin, Erythromycin, and Trimethoprim-sulfamethoxazole contrasts with the results reported by Mukta and Manir (2016) and Islam *et al.* (2010), while being consistent with the findings of Sileshi and Munees (2016) and Martha *et al*. (2016).

Isolates of *Staphylococcus aureus* demonstrated complete susceptibility to Ciprofloxacin (100%) and significant sensitivity to Gentamycin, Vancomycin, and Erythromycin (83.33%), as well as Chloramphenicol and Tetracycline (71.4%). However, these isolates showed resistance to Cephoxitin (88.09%) and Penicillin (71.4%). The high levels of resistance to Cephoxitin and Penicillin in this study support the findings of Thaker *et al.* (2013) in India, where *Staphylococcus aureus* isolates exhibited complete resistance to Penicillin-G.

Salmonella Species isolates were found to be susceptible to Ciprofloxacin (100%), Gentamycin (86.7%), Chloramphenicol (83.3%), and Erythromycin (66.9%) and Trimthoprim-sulfamethoxazole (66.7%). However, there was resistance observed with Tetracycline (86.7%), Vancomycin (83.3%), Penicillin (76.7%) and Cephoxitin (70.0%). The high resistances of these antibiotics are similar to the study conducted by Makwin*et al*. (2014) in Keffi town and the susceptibility of some of these antibiotics to *Salmonella* Species is similar to the work conducted by Sileshi and Munees (2016), Martha *et al*. (2016) and Mukta and Manir (2016).

Pseudomonas aerugunosa was highly susceptible to Ciprofloxacin (100%), Chloramphenicol (87.5%), Erythromycin (75.0%), and Gentamycin (62.5%). However, there was high resistance in Vancomycin and Penicillin (100%) and Cephoxitin (68.75%), while *Bacillus species* was highly susceptible to Ciprofloxacin and Gentamycin (90.90%), Chloramphenicol (86.4%) and Erythromycin (81.82%) and Trimethoprim-sulfamethoxazole (90.90%). However, there was also some high resistance of *Bacillus species* isolates to Cephoxitin (90.91%), Vancomycin (81.82%), Penicillin (86.4%) and Tetracycline (77.3%). *Proteus Species* were highly susceptible to Ciprofloxacin (100%), Chloramphenicol (87.5%), Gentamycin (75.0%), Erythromycin (68.75%) and Trimethoprim-sulfamethoxazole (75.0%). However, the *Proteus Species* isolates were highly resistant to Vancomycin, Penicillin, Cephoxitin and Tetracycline (75.0%). *Streptococcus species* were highly susceptible to Ciprofloxacin, Gentamycin, and Chloramphenicol (90.0%), Trimethoprim-sulfamethoxazole (80.0%) and Erythromycin (70.0%). However, some of the *Streptococcus species* isolates were highly resistant to Penicillin, Cephoxitin and Vancomycin (90.0%) and Tetracycline (85.0%). The sensitivity pattern of these bacteria isolates isolated is comparable to the reports of earlier researchers Inyang (2009), Udo *et al.* (2001), Tagoe *et al.* (2011), Makut*et al.* (2013), Makwin*et al.* (2014), Muktar and Manir (2016), Sileshi and Munees (2016) and Martha *et al.* (2016). All the isolates were susceptible to Ciprofloxacin and Gentamycin and resistance to Penicillin and Cephoxitin which is in agreement with the findings by Idress *et al*. (2014), Belayneh *et al.* (2018) and Martha *et al.* (2016).

The occurrence of antibiotic-resistant strains of *Echerichia coli, S. aureus, Salmonella species, P. aeruginosa*, *Bacillus species*, *Proteus species*, and *Streptococcus species* in unpasteurized milk highlights the consequences of both the appropriate and inappropriate use of antibiotics within society. This situation is particularly concerning given the widespread and indiscriminate use of antibiotics among the Nigerian population, including those in Makurdi. The public health implications of this study suggest that antimicrobial-resistant strains of pathogenic bacteria could potentially infect humans through the consumption of contaminated unpasteurized milk sourced from selected markets in the Makurdi area, which may ultimately result in treatment failures for individuals consuming this milk.

5. CONCLUSION

In the Makurdi metropolis, unpasteurized milk sold in various markets exhibited higher levels of pathogenic microorganisms. The bacterial isolates identified in this study are believed to have contaminated the milk samples from multiple sources, likely due to inadequate handling and storage practices during milk collection. Factors contributing to this contamination include the environment, the cleanliness of utensils, the hygiene of the animals providing the milk, and the sanitary conditions maintained by the milk collectors. The research findings indicate that Ciprofloxacin demonstrated a remarkable susceptibility to the isolates, achieving 100% efficacy, a fact that was previously unknown. In contrast, Vancomycin and Penicillin exhibited complete resistance.This study also provides insights into the sensitivity profiles of *Echerichia coli*, *Staphylococcusaureus*, *Salmonella* species, *Pseudomonas aeruginosa*, *Bacillus* species, *Proteus* species, and *Streptococcus* species isolated from unpasteurized milk samples, tested against nine commonly used antibiotics in Makurdi. Additionally, it highlights the issue of multi-drug resistance among certain bacterial isolates in these milk samples, which raises concerns about the indiscriminate use of antibiotics in the region. This situation poses significant risks to human health, as a considerable portion of the population consumes this milk. Furthermore, these multi-drug resistant bacteria may become untreatable with standard therapeutic drugs and have the potential to transfer their resistance genes to other bacterial species**.**

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