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Research Article

Antimicrobial Resistance Profiles of *Salmonella* spp. Isolated from Clinical Samples in Niamey, Niger

Alio Mahamadou Fody[1, 2, 3, *], Chaibou Yaou[3], Hadiza Ibrahim Bawa[4], Laouali Boubou[5], Mohamed Alassane Halawen[6], Ramatou Sidikou[7], Hassimi Sadou[3], Nicolas Barro[2]

[1]Department of Life and Earth Sciences, Abdou Moumouni University, Niamey, Niger
[2]Department of Life and Earth Sciences, Joseph Ki-Zerbo University, Ouagadougou, Burkina Faso
[3]Department of Chemistry, Abdou Moumouni University, Niamey, Niger
[4]Department of Life and Earth Sciences, Ledea Bernard Ouedraogo University, Ouahigouya, Burkina Faso
[5]Bacteriology Laboratory of Niamey National Hospital (NNH), Niamey, Niger
[6]Microbiology Laboratory of the General Reference Hospital (GRH), Niamey, Niger
[7]Department of Biology, Abdou Moumouni University, Niamey, Niger

Abstract

Background: *Salmonella* spp. are leading causes of gastroenteritis and typhoid fever worldwide, with hospital-acquired infections posing significant clinical challenges. The emergence of antibiotic resistance among clinical isolates has been increasingly recognized as a major contributor to treatment failure in affected patients. Objective: This study aimed to determine the prevalence and antimicrobial resistance patterns of *Salmonella* spp. isolated from clinical specimens at the National Hospital of Niamey, Niger. Method: Stool and blood samples were collected from patients presenting with clinical signs of enteric fever or gastroenteritis. *Salmonella* spp. was identified via standard microbiological methods, and antimicrobial susceptibility testing was performed via the disk diffusion method on Mueller–Hinton agar in accordance with Clinical and Laboratory Standards Institute (CLSI) guidelines. Extended-spectrum beta-lactamase (ESBL) production was confirmed by a double-disk synergy test, which assessed the enhancement of inhibition zones between amoxicillin/clavulanic acid and third-generation cephalosporins. Results: Seventy-seven *Salmonella* isolates were recovered, yielding an overall prevalence of 2.55%. The majority of the isolates (80.52%) were obtained from children under 5 years of age. High resistance rates were observed against amoxicillin and amoxicillin/clavulanic acid (79% each). Multidrug resistance (resistance to ≥3 antibiotic classes) was detected in 25.97% of the isolates, whereas 35% were resistant to at least two antibiotics. Notably, 5.19% of the isolates were confirmed as ESBL-producing strains. Conclusion: These findings highlight the significant burden of multidrug-resistant *Salmonella* spp., including ESBL producers, in the clinical setting of Niamey. The high prevalence of resistance to commonly used antibiotics underscores the urgent need for enhanced antimicrobial resistance surveillance, rational antibiotic use, and infection control strategies to curb the spread of resistant strains in healthcare and community environments.

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[*]Correspondence: Alio Mahamadou Fody (juniorfodym@gmail.com)

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Keywords

Salmonella spp., Clinical Isolates, Antimicrobial Resistance, Multidrug Resistance, Extended-spectrum Beta-lactamases (ESBL), Niger

1. Introduction

Salmonella is a gram-negative enteric bacterium responsible for a significant burden of foodborne illness in both humans and animals worldwide [1, 2]. In 2015, according to Shukee et *al.*, the genus encompasses a diverse group of pathogens that collectively account for an estimated 93.8 million annual cases of gastroenteritis worldwide, resulting in approximately 155,000 deaths [3]. According to WHO (2018), *Salmonella enterica* serovars Typhi and Paratyphi are the causative agents of enteric fever, contributing to 11–21 million cases of typhoid fever and 128,000–161,000 associated deaths annually, as well as an estimated 6 million cases of paratyphoid fever, with over 54,000 fatalities [4]. In sub-Saharan Africa, nontyphoidal *Salmonella* infections are a major cause of invasive bacterial disease, contributing to approximately 680,000 deaths each year according to Post et *al.*, in 2029 [5].

The emergence and spread of antimicrobial resistance (AMR) in *Salmonella* spp. represent critical public health challenges. WHO declared 3GC-resistant *Enterobacteria* & FQ-resistant *Salmonella* are critical and high group pathogen [4]. Resistance to first- and second-line antibiotics compromises the efficacy of clinical treatment, leading to prolonged illness, increased risk of complications, and increased mortality rates [2]. In sub-Saharan West Africa, the mortality burden attributable to AMR is particularly high, with estimates reaching 27.3 deaths per 100,000 infections [6]. Furthermore, the World Bank projects that AMR could increase healthcare expenditures by up to 25% in low-income countries and by 8% globally [7].

Multidrug-resistant (MDR) *Salmonella* strains have been increasingly reported during outbreaks, undermining therapeutic options for salmonellosis [8]. In Burkina Faso, MDR phenotypes are increasing among *Salmonella* isolates from pediatric populations [9], whereas in Niger, recent studies have documented the emergence of resistance to third-generation cephalosporins and fluoroquinolone key agents used in the management of invasive salmonellosis and shigellosis, highlighting the need for cautious antibiotic use and continuous surveillance [10].

This study aimed to evaluate the antimicrobial susceptibility profiles of *Salmonella* spp. isolated from blood and stool samples at the National Hospital of Niamey, Niger. These findings provide critical insights into the current patterns of resistance to antibiotics commonly employed in the treatment of gastroenteritis and enteric fever, informing clinical decision-making and public health interventions in the region.

2. Materials and Methods

2.1. Study Design and Duration

A descriptive cross-sectional study was conducted between July 2017 and May 2018 at the Laboratory of Clinical Biology, National Hospital of Niamey. Stool and blood samples were collected from patients who presented with diarrhea or clinically suspected typhoid fever.

2.2. Sample Collection and Culture Procedures

2.2.1. Stool Samples

Stool samples were collected aseptically in sterile containers. Approximately one gram of each sample was suspended in physiological saline for pre-enrichment. A few drops of the suspension were subsequently inoculated into selenite broth and incubated at 37 ℃ for 10 hours for selective enrichment. Following enrichment, the cultures were streaked onto Hektoen enteric agar and incubated at 37 ℃ for 24 hours for isolation of *Salmonella* species according to FDA [11].

2.2.2. Blood Samples

Blood samples were collected directly into aerobic blood culture bottles and incubated in the bioMerieux Bact/Alert 3D automated culture system at 37 ℃ for up to 5 days [12]. Bottles flagged as positive by the system were subcultured by inoculating 10 µL onto Blood Agar and Hektoen Enteric Agar [or specify appropriate media], followed by incubation at 37 ℃ for 18–24 hours [12].

2.3. Identification of Isolates

Suspected *Salmonella* colonies were identified by their morphological characteristics on selective media, cultural properties, and standard biochemical tests, including oxidase assays (negative for *Salmonella*). Presumptive isolates were further confirmed via the API 20E system (bioMerieux), a standardized, miniaturized panel of 20 biochemical tests designed for the identification of *Enterobacteriaceae* and other gram-negative bacilli [13]. Identification was achieved through numerical code generation and comparison with the reference database.

2.4. Antibiotic Susceptibility Testing

Antibiotic susceptibility was assessed via the disk diffusion method on Mueller–Hinton agar (Liofilchem, Italy) in accordance with the guidelines of the *Antibiogram Committee of the French Society for Microbiology* (CA-SFM) [14]. The tests included antibiotics from three major classes: be-ta-lactams (amoxicillin [20 µg], amoxicillin-clavulanic acid [20 µg + 10 µg], ceftriaxone [30 µg], ceftazidime [10 µg], cefotaxime [5 µg], and imipenem [10 µg]), aminoglycosides (gentamicin [10 µg] and amikacin [30 µg]), and quinolones (nalidixic acid [30 µg] and ciprofloxacin [5 µg]).

Phenotypic detection of extended-spectrum beta-lactamase (ESBL) production was performed via the double-disk synergy test (DDST), with ceftriaxone (30 µg) and cefotaxime (5 µg) disks placed at a distance of 20 mm (edge-to-edge) from an amoxicillin–clavulanic acid (30 µg) disk, as recommended by the CA-SFM 2017 guidelines [14]. A positive result was defined as an increase in the inhibition zone toward the clavulanic acid disk, indicating synergy.

2.5. Ethical Considerations

All isolates were derived from biological specimens collected during routine clinical procedures from patients, with ethical approval granted by the Ethics Committee of the National Hospital of Niamey (Authorization No. 00075/DHNN/DAF/SGRH, dated November 15, 2016). No additional sampling was performed for research purposes.

2.6. Statistical Analysis

The data were entered and analyzed via Microsoft Excel (Microsoft® Office 2019). The chi-square test was used to evaluate associations between categorical variables. A p-value < 0.05 was considered statistically significant.

3. Results

A total of 3,024 clinical specimens, comprising 2,837 stool samples and 187 blood culture samples, were analyzed at the Bacteriology Unit of the National Hospital of Niamey during the study period.

3.1. Prevalence of *Salmonella* spp. by Sample Type

Overall, 77 *Salmonella* spp. isolates were identified, resulting in a total prevalence of 2.55% (95% CI: 2.02–3.17%). The distribution of *Salmonella* isolates by specimen type is summarized in Table 1.

Among the stool samples, 67 out of 2,837 tested positive for *Salmonella* spp., yielding a prevalence of 2.22% (95% CI: 1.73–2.81%). Among the blood cultures, 10 out of the 187 samples were positive, corresponding to a prevalence of 5.35% (95% CI: 2.68–9.42%). This difference in prevalence between stool and blood samples was statistically significant ($\chi^2 = 13.78$, $p < 0.001$), indicating that a greater proportion of *Salmonella* was detected in blood cultures despite the smaller sample size.

Table 1*. Prevalence of Salmonella* spp. *by sample type.*

Sample types	Sample numbers	*Salmonella* spp. prevalence% (n)
Stool samples	2,837	2.22 (60)
Blood samples	187	5.35 (10)
Total	3,024	2.55 (77)

3.2. Prevalence of *Salmonella* spp. Strains According to Sociodemographic Characteristics

The prevalence of *Salmonella* spp. was 53.24% (n = 41) among male patients and 46.76% (n = 36) among female patients. Statistical analysis revealed no significant difference in the prevalence of *Salmonella* between the sexes (p = 0.5485).

Salmonellosis affects individuals across all age groups; however, a marked variation in prevalence was observed according to age (Figure 1). The highest prevalence was recorded in children aged 0–5 years, with 80.52% (n = 62) of the cases. In contrast, the prevalence was substantially lower in the older age groups: 3.90% (n = 3) in the 6–18 years group, 7.79% (n = 6) in the 19–25 years group, 2.60% (n = 2) in the 26–45 years group, and 5.19% (n = 4) in the 46–60 years group. This age-related difference in *Salmonella* spp. prevalence was statistically significant (p < 0.0001).

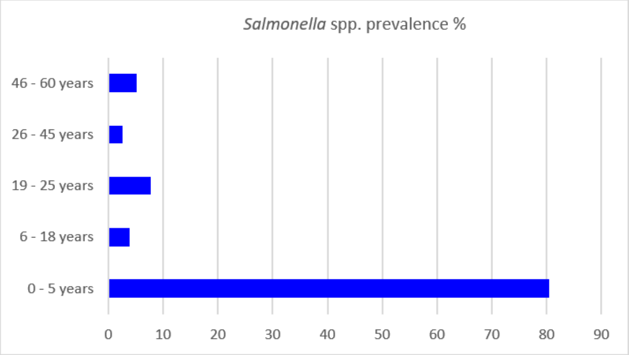

Figure 1. *Prevalence of Salmonella spp. by age (years).*

3.3. Prevalence of *Salmonella* spp. Isolates by Sample Origin

Analysis of the data presented in Table 2 revealed a *Salmonella* spp. prevalence of 40.26% among outpatients, which was significantly higher ($p < 0.0001$) than that among inpatients across hospitalization departments. Among inpatient clinics, the highest prevalence was observed in the pediatric referral clinic (31.17%), followed by the Pediatrics A clinic (23.38%). Lower prevalence rates were recorded in the Medicine B clinic (3.90%) and the Pediatrics B clinic (1.30%). A statistically significant variation in Salmonella spp. prevalence was also observed across different hospitalization departments ($p < 0.0001$).

Table 2. *Prevalence of Salmonella* spp. *isolates according to sample origin.*

Samples origin	*Salmonella* spp. prevalence% (n)
Pediatrics A office	23.38 (18)
Pediatrics B office	1.30 (1)
Pediatric referral office	31.17 (24)
Medicine B office	3.90 (3)
External	40.26 (31)

3.4. Antibiotic Susceptibility of *Salmonella* spp. Isolates

Analysis of *Salmonella* spp. Isolates obtained from blood cultures (Figure 2) revealed high resistance rates within the beta-lactam antibiotic class: 90% of the isolates were resistant to both amoxicillin and amoxicillin/clavulanic acid. In contrast, susceptibility to third-generation cephalosporins was high, with 90% of the isolates showing sensitivity to ceftazidime, cefotaxime, and ceftriaxone. All the isolates were fully susceptible (100% sensitivity) to the carbapenem imipenem. Among the 10 antibiotics tested, only a single isolate exhibited multidrug resistance, defined as resistance to eight antimicrobial agents.

With respect to aminoglycosides, 90% of the isolates were susceptible to gentamicin, whereas 100% were susceptible to amikacin. Within the quinolone class, both nalidixic acid and ciprofloxacin demonstrated 90% susceptibility rates.

These findings highlight a high level of resistance to certain beta-lactams but preserved susceptibility to extended-spectrum cephalosporins, carbapenems, aminoglycosides, and fluoroquinolones among the majority of *Salmonella* spp. Isolates.

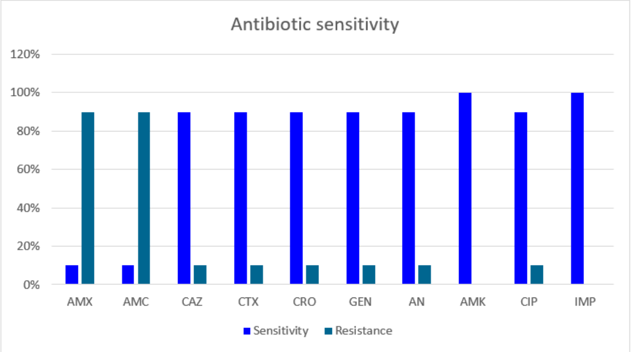

Figure 2. *Antibiotic susceptibility of Salmonella* spp. *Isolated from blood samples.*

Analysis of stool-derived bacterial isolates (Figure 3) revealed antimicrobial susceptibility patterns across key antibiotic classes. Among β-lactam agents, amoxicillin and amoxicillin-clavulanate (augmentin) have low susceptibility rates, with a reported sensitivity of 31%. In contrast, extended-spectrum cephalosporins and carbapenems demonstrated greater efficacy: ceftazidime had 60% sensitivity, cefotaxime had 67%, ceftriaxone had 70%, and imipenem had a markedly high sensitivity rate of 99%.

Within the aminoglycoside class, the susceptibility rates were 64% for gentamicin and 94% for amikacin, indicating the superior activity of the latter. Specifically, among *Salmonella* spp. Isolates, susceptibility to nalidixic acid was 73%, whereas 75% of isolates were susceptible to amikacin.

These findings highlight high resistance to penicillins, variable sensitivity to cephalosporins, and preserved susceptibility to carbapenems and amikacin, suggesting that these agents may remain effective therapeutic options in the management of infections caused by these enteric pathogens.

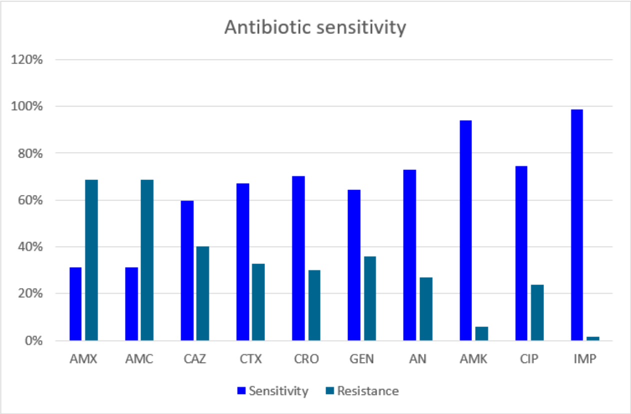

***Figure 3**. Antibiotic susceptibility of Salmonella spp. Isolated from stool samples.*

3.5. Prevalence of Multidrug-resistant *Salmonella* spp. Strains

Analysis of the data presented in Table 3 revealed that 20.78% of the *Salmonella* spp. isolates were susceptible to all antibiotics tested in this study. In contrast, only a single isolate (1.30%), obtained from a stool sample, exhibited resistance to all the tested antimicrobial agents. Resistance to nine, eight, seven, six, and five antibiotics was observed in 2.60%, 12.99%, 3.90%, 3.90%, and 1.30% of the isolates, respectively. Notably, a substantial proportion of the isolates (35.06%) displayed resistance to two antibiotics, indicating a high prevalence of multidrug resistance among the *Salmonella* strains analyzed.

***Table 3**. Prevalence of multidrug-resistant Salmonella* spp.

Number of resistant antibiotics	Resistance of *Salmonella* spp% (n)
10	1.30 (1)
9	2.60 (2)
8	12.99 (10)
7	3.90 (3)
6	3.90 (3)
5	1.30 (1)
4	2.60 (2)
3	9.09 (7)
2	35.06 (27)
1	6.49 (5)

3.6. Prevalence of Extended-spectrum Beta-lactamase (ESBL)-producing *Salmonella* spp.

An analysis of the data in Table 4 revealed that 20 *Salmonella* spp. isolates were resistant to at least five antibiotics, resulting in a multidrug resistance prevalence of 25.97%. Among these multidrug-resistant isolates, four were identified as extended-spectrum beta-lactamase (ESBL) producers and were isolated from stool samples. This corresponds to an overall prevalence of 5.19% for ESBL-producing *Salmonella* spp. (Table 4).

***Table 4**. Prevalence of ESBL-producing Salmonella spp.*

Samples types	MDR *Salmonella* spp.	*Salmonella* spp. ESBL
Stool samples	24.67% (19)	5.19% (4)
Blood samples	1.30% (1)	0.00% (0)
Total	25.97% (20)	5.19% (4)

4. Discussion

Salmonella infections represent a significant public health burden in Africa, particularly among poor urban populations, where they contribute to a disproportionate number of deaths [15]. This burden is exacerbated in low-income countries because of inadequate control measures and limited adherence to proper food safety practices [16].

In this study, the prevalence of *Salmonella* spp. in clinical stool samples was 2.22%. This finding is comparable to reports from Burkina Faso (3.07%) and Nepal (3.1%) ($p = 0.6547$) [17, 18]. However, it is significantly lower than the prevalence reported in other studies conducted in Burkina Faso (9.50%), Tanzania (16.5%), and Iraq (14.8%) ($p = 0.0068$) [9, 19, 20]. These discrepancies may be attributed to differences in study design, sample size, population characteristics, and regional variations in hygiene, sanitation, and access to healthcare.

Notably, a low prevalence of 0.33% was observed in the blood culture samples. This result is significantly lower (p <

0.0001) than those reported in studies from Kenya (3.54%), the Democratic Republic of the Congo (41.66%), Nigeria (7.27%), and Nepal (3.10%) [15, 21-23]. The low bacteremia rate in our study may be partly explained by the relatively short duration of the study period and the limited sample size, which could affect the detection of bloodstream infections. The observed disparity suggests a greater clinical severity associated with bacteremia *Salmonella* infections in this population, although the lower number of blood samples limits direct comparison. These findings highlight the importance of blood culture surveillance in the diagnosis of invasive salmonellosis.

With respect to sex distribution, *Salmonella* spp. was more frequently isolated from male patients (53.24%) than from female patients (46.76%). This observation aligns with findings from studies in Mali [25] and Tanzania [26]. The higher prevalence among males may be associated with behavioural and socioeconomic factors, such as a greater frequency of eating outside the home, potentially increasing exposure to contaminated food or water sources.

Children aged 0–5 years presented the highest prevalence of *Salmonella* spp. infection (80.52%). This finding is consistent with data from Iran, where a high incidence was reported in children under 4 years of age [27], and from Bangladesh, where 62% of typhoid fever cases were attributed to *Salmonella* spp. [28]. The heightened susceptibility of young children may be linked to widespread malnutrition in resource-limited settings, which can compromise intestinal immunity and alter the gut microbiota, thereby increasing vulnerability to enteric pathogens [29].

Antimicrobial susceptibility testing revealed high levels of resistance to amoxicillin and amoxicillin/clavulanic acid. These findings are consistent with a study in Nigeria, which reported 100% resistance to amoxicillin/clavulanic acid [22]. In contrast, studies in Nepal and Kathmandu demonstrated high sensitivity (100% and 91%, respectively) to amoxicillin [18, 23]. The observed variation in resistance patterns may reflect differences in antibiotic usage practices, including widespread self-medication for symptoms such as fever, diarrhea, and respiratory infections, as well as the empirical prescription of antibiotics without microbiological confirmation or susceptibility testing [28].

Resistance to third-generation cephalosporins ceftazidime, cefotaxime, and ceftriaxone was low in our isolates. These results are in agreement with previous reports from Nepal [18] and Niger [10]. However, a study in Nigeria reported 100% resistance to ceftazidime and cefotaxime [22], which may be attributed to prior antibiotic exposure, as the isolates in that study were obtained from patients already experiencing febrile illness, likely after self-treatment.

Our isolates showed high susceptibility to amikacin (97%) and imipenem (99%), indicating that these agents remain effective therapeutic options for severe salmonellosis. A study in Cameroon reported 90% susceptibility to imipenem [24]; the slightly lower sensitivity may reflect greater -carbapenem use in that setting, potentially driving selective pressure.

Multidrug resistance (MDR), defined as resistance to two or more antibiotic classes, was observed in 35% of *Salmonella* spp. isolates. This is lower than the 85% MDR prevalence reported in a Bangladeshi study based on blood isolates [28], a difference that may be influenced by variations in sample size and patient population. Notably, 13% of the isolates in our study exhibited resistance to at least eight different antibiotics, meeting the criteria for extensive drug resistance. This finding is concerning and aligns with reports from South Africa, Ethiopia and Canada, where 22% of isolates were resistant to three or more antibiotics, 31% to five antibiotics, and 59% to three different antibiotic classes [30-32]. These data underscore a growing trend of increasing antimicrobial resistance among clinical *Salmonella* strains.

Among the MDR isolates, 4 (5.19%) were confirmed as extended-spectrum beta-lactamase (ESBL)-producing strains. This prevalence is comparable to reports from Canada (2%) and Thailand (5.1%) [32, 33] but lower than rates reported in Nigeria (40.7%) and Qatar (10%) among patients with confirmed typhoid fever [34, 35]. The higher ESBL rates in the latter studies may reflect the clinical context—specifically, that isolates were obtained from patients with established enteric fever who had likely received prior antimicrobial treatment, thereby selecting for resistant strains.

In conclusion, our findings highlight the persistent burden of *Salmonella* infections, particularly among young children and males, and underscore the growing challenge of antimicrobial resistance. Continued surveillance, improved diagnostic practices, rational antibiotic use, and public health interventions targeting food and water safety are essential to mitigate the impact of *Salmonella* infections in low-resource settings.

5. Conclusion

This study, which was conducted over 10 months at the National Hospital of Niamey, allowed the isolation and identification of *Salmonella* spp. strains from clinical stool and blood samples. The results revealed a greater prevalence of Salmonella spp. in stool samples than in blood samples. Most of the isolated *Salmonella* strains were sensitive to the majority of the tested antibiotics, but some showed resistance to certain drugs. Multiple drug-resistant Salmonella strains have been identified, including some that produce extended-spectrum beta-lactamases (ESBLs). Therefore, it is essential to establish a national surveillance system as soon as possible for ESBL-producing Salmonella strains to help prevent their spread.

Abbreviations

AMR	Antimicrobial Resistance
MDR	Multidrug Resistance
ESBL	Extended-Spectrum Beta-Lactamase

CLSI	Clinical and Laboratory Standards Institute
CA-SFM	Antibiogram Committee of the French Society for Microbiology
DDST	Double-Disk Synergy Test
API 20E	Analytical Profile Index 20 Enterobacteriaceae
SFM	French Society for Microbiology
EUCAST	European Committee on Antimicrobial Susceptibility Testing
LaBESTA	Laboratory of Molecular Biology, Epidemiology and Surveillance of Foodborne Bacteria and Viruses
NNH	Niamey National Hospital
GRH	General Reference Hospital

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Author Contributions

Alio Mahamadou Fody: Conceptualization, Methodology, Writing – original draft, Writing – review & editing

Chaibou Yaou: Writing – review & editing

Hadiza Ibrahim Bawa: Writing – review & editing

Laouali Boubou: Data curation, Supervision

Mohamed Alassane Halawen: Writing – review & editing

Ramatou Sidikou: Visualization

Hassimi Sadou: Visualization

Nicolas Barro: Visualization

Data Availability Statement

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Conflicts of Interest

The authors have not declared any conflicts of interest.

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