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## Antibiotic Residues in fresh Chicken, Fish and Beef Meat samples in Maiduguri, Nigeria

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**ABSTRACT**

Prudent use of antibiotics in animal industry is essential for treatment of diseases and preventive measure. However, uncontrolled use of these agents in large amounts and consistently could result in the formation of antibiotic residues in muscle and organs of animal. This study aimed to determine the occurrence of antibiotic residue in fresh meats in Maiduguri. A cross-sectional study was conducted from September 2022 to December 2022 to determine the antibiotic residue in 3 sources of meat samples. A total of 150 fresh meat samples (chicken, fish and beef) were collected from different sale points Maiduguri, and analyzed using a Premi® Test kit (R-Biopharm AG, Germany), a microbial screening commercial test kit for the detection of antibiotic residues. The results showed an overall antibiotic residue of 127/150 (84.7%) meat samples were positive for antibiotic residues. High antibiotic residues were found in Chicken meats 47/50 (94%), followed by fish 43/50 (86%), and beef 37/50 (74%). The results in this study show the presence of antibiotic residues in meat. Therefore, it is recommended that effective legislation on antibiotic use in animals should be enforced.

**Keywords:** Antibiotic residues; Antibiotic resistance; Food safety; Public health; Meat

**INTRODUCTION**

The quest for meat is increasing globally, leading to the use of antimicrobials in livestock production to increase productivity and treat diseases (da Silva, 2019). This has resulted in the consumption of antimicrobials in food animal production, which has been projected to increase by 67% by 2030 (Okocha *et al.*, 2018; Lee *et al.*, 2018). Currently, the use of antibiotics in animal husbandry is not limited to therapeutic purposes but is mostly used for prophylactic purposes and as growth promoters to achieve high return in protein (Dipeolu and Alonge, 2002). Antibiotics have been used for treating bacterial infections in both humans and animals; however, their application in animals has doubled in recent years (Aarestrup, 2012; Bacanl and Başaran, 2019; Arsene *et al.*, 2021). In a particular report, the global consumption of antibiotics in livestock farming is reported to be over 80% and mostly not for therapeutic purposes (Boeckel *et al.*, 2015). The most commonly used antibiotics include tetracyclines, which amount to 70% (Page and Gantier, 2014). The presence of drug residues in meat above the maximum residue limit (MRL), especially when withdrawal periods are not observed, has become a global issue (Kantati, 2011; Liu *et al.*, 2020). The lack of observation of withdrawal periods before

processing animals for consumption is recognised worldwide (Adesokan *et al.*, 2013; Ezenduka *et al.*, 2014).

The emergence of antibiotic resistance has been driven by many factors including misuse and overuse of antibiotics for therapeutic and non-therapeutic purposes. The problem of antibiotic resistance (ABR) has a negative impact on the health and socio-economic status of the people (Ferri *et al.*, 2017). The World Health Organization (WHO) expressed concern on the dissemination of antibiotic residues in food chains due to excessive use of antibiotics in farming and animal husbandry. The practice of uncontrolled use of antibiotics exposes humans to ABR food-borne pathogens and commensals through animal food products and the environment (Silbergeld *et al.*, 2008; Cháfer-Pericás *et al.*, 2010; Abdullahi *et al.*, 2015, Mustapha *et al.*, 2021). Food contamination has been a major source of infection, especially in food products, poultry products, and seafood products (Akbar and Anal, 2011; Cunha, 2001; Jans *et al.*, 2018; Sugrue *et al.*, 2019). The excessively used antibiotics in animal husbandry could be deposited as residues in animal tissues and the effect could be on several fronts such as development of resistance by intestinal microbiota, allergies, carcinogenicity, mutagenicity, teratogenicity, nephropathy, immunological disorders, hepatotoxicity, reproductive disorders and transfer of mobile resistance genes

(Aryal, 2001; Alvarez-Fernandez *et al.*, 2013; Federal Ministries of Agriculture, Environment and Health, 2017; Panigrahi *et al.*, 2017). There is growing evidence of antibiotic residues in Nigeria, particularly in meat and milk from animal source. A study conducted to assess the antibiotic residues in raw meat sold in six abattoirs in Kwara State, Nigeria found antibiotic residues above recommended permissible levels (Olasoju *et al.*, 2021). Another study examined the presence of antibiotic, oxytetracycline in Lagos and Ogun states, Southwestern Nigeria, and indicated presence of antibiotic residues meat above recommended levels (Olumayowa *et al.*, 2021). In another study in Abuja, results of antimicrobial residues in beef indicated 89.3% from cattle samples revealed occurrence of antibiotic residues (Omeiza *et al.*, 2012). Different antibiotic residues from raw meat were detected in Kaduna, Northwestern Nigeria and reported 67% (4/18) from meat sold in six slaughter houses (Lateefat *et al.*, 2022). Different are used in conducting antimicrobial residue studies to assess the presence of antimicrobial residue in various products (Oloso *et al.*, 2018).

In Nigeria, prudence in the use of antibiotics in both human and animal use is lacking due to the lack of proper antibiotic use and enforcement of the existing laws by agencies (Dipeolu and Alonge, 2002). The antibiotic residues in meat and meat products sold in Maiduguri are not yet ascertained, and studies regarding the antibiotic residues in meat from this sub-region are scarce. Therefore, this study aimed to screen antibiotics residues in meats from different meat sources in Maiduguri, Borno State.

## MATERIALS AND METHODS

### Study Area

The study was conducted in Maiduguri, Borno State, Nigeria from September 2022 to December 2022. Maiduguri is located in the North-Eastern part of Nigeria which lies within latitude 11.15°N and longitude 30.05°E in the Sudano-Sahelian savanna zone with a dense population who are mostly crop farmers, fishermen, herdsman and traders (Udo, 1978). The state has an area of 71,210sq km with the population of 4,151,193 according to the National census conducted in 2006 (NPC, 2006).

### Sample Collection

A total sample of 150 meats (broiler chickens n=50, beef=50, and fish, n=50) were purchased from different sale points in Maiduguri Metropolitan of Borno State, Nigeria.

### Study Design

A cross-sectional study was conducted from September 2022 to December 2022 to determine the antibiotic residue in 3 sources of meat samples. Different parts of the chickens including kidney, liver, gizzard and beef and fish muscles were harvested. These parts are reported to have high concentration of antibiotics (Alvarez-Fernandez *et al.*, 2013). The meats parts were bought and brought to the Microbiology Department of University of Maiduguri for analysis.

### Sample Preparation

The meat samples were cut into pieces of approximately 2cm<sup>3</sup> separately and each set were put into the meat-press. The

pressure was increased slowly and held constant until approximately 200µl of meat-juice was obtained.

### Antibiotic Residues Detection

Premi® Test kit (R-Biopharm AG, Germany) a microbial screening commercial test kit was used for the qualitative detection of antibiotic residues. Premi® Test kit is a rapid test kit based on principle of inhibition of microorganisms. It comes with ampoules of agar imbedded with standardized number of spores of *Bacillus stearothermophilus* as test organism and Bromocrescol purple colour indicator. The Premi test is considered rapid, easy to use and suitable as a preliminary screening test for antibiotic residues with specificity of 95.3% and sensitivity of 72.5%. This method is mainly employed for the preliminary study to assess the presence of antibiotic residues.

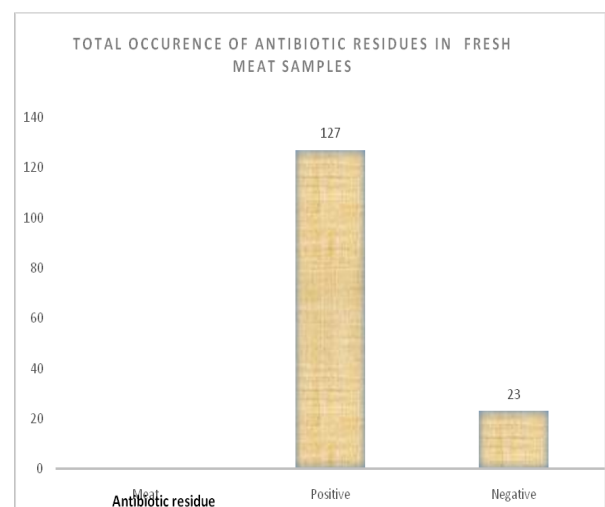
Gently, 100µL of the meat-juice was pipetted into the ampoule without distortion of the agar and then covered with perforated foil based on Manufacturer's specifications. The ampoules were incubated for 20 minutes at room temperature, after which it was incubated inside the water bath at 65 °C for 3 hours. Indication of the results of samples was recorded when the negative control shows a clear colour change to yellow after 3 hours of incubation while the ampoules that remained purple after the incubation were recorded as positive for antimicrobial residues. Data obtained was analysed using DATA Tab Team (2023). Chi-square analysis was conducted and value (p<0.05) was considered significant.

### Ethical Statement

Consent was duly obtained from the sale points and approval was given before the specimens were collected for further investigations in the laboratory.

## RESULTS AND DISCUSSION

The occurrence of antibiotic residues in fresh meat samples in Maiduguri, Nigeria was determined. Out of the 150 meat samples (chickens, n=50, beef=50, and fish, n=50) collected and examined, using Premi® Test kit. In this study, different meat samples were sampled to detect the presence of antibiotic residues, a total/overall prevalence of antibiotic residues was found to be high 127/150 (84.7%) (Figure 1).



**Figure 1:** A total occurrence of antibiotic residues in fresh meat samples in Maiduguri, Nigeria.

A significant association was observed between meat samples and the occurrence of antibiotic residues,  $\chi^2$  (2, N=150,) = 7.8055,  $p=0.0202$ . In a similar study, Ekene (2019) found a strong association between the occurrence of antimicrobial residues and the meat type ( $\chi^2$  value = 64.5,  $p$  value < 0.0001). The reason behind this may be due to exposure of the animals to antibiotics. Contrary to the current study, Onwumere-Idolor *et al.* (2021) reported no significant association between meat type and antibiotic residues ( $\chi^2=5.206$ ,  $p=0.074$ ). This difference could be explained by the detection method which was employed (The Four Plate Test) by Onwumere-Idolor *et al.* (2021).

The current study revealed high antibiotic residues this might be due to use of uncontrolled use of antibiotics in the study area. In particular, Chicken meat contained highest antibiotic residues 47/50 (94%), and the result is found to be consisted with a Lebanese study that found 77.5% of 80 samples of chicken meat were contaminated with antibiotic residues (Adla and Nada 2019). In contrast to a study of contamination of antibiotic residues in Portugal on 92 samples of chicken and reported 42% (Pena *et al.*, 2010). The difference in the occurrence might be attributed to the detection methods or controlled use of antibiotics. High levels of antibiotic residues were reported in other parts of Nigeria. Ezenduka *et al.*, (2019) reported 64% (155/400) of chicken meat to have antibiotic residues, another study demonstrated high antibiotic residues in chicken meat in South Western Nigeria (Onipede *et al.*, 2021), and both are lower than the present study. This could be due to high animal husbandry in northern Nigeria and excessive use of antibiotics. Conversely, antibiotic residues in poultry meat were reported to be lower in European countries, the report confirmed only 15% of antibiotic residues from different countries. The disparity of antibiotic residues burden among countries is account for the use and misuse of antibiotics and strict regulations in the countries. In Nigeria for example, the use of antibiotic in animal husbandry by farmers is common for clinical treatment and prophylaxis purposes.

The antibiotic residue in fish meat in the current study is 86% (43/50) (Table 1). The antibiotic residues burden in fish in this study was similar to that obtained by various studies; Barani and Falla (2014) in Iran, Olatoye and Basiru (2013) in Nigeria, and 26.9% (28/104) in Vietnam by Pham *et al.*, (2015). Chicken meat contained high antibiotic residues 47/50 (94%), followed by fish 43/50 (86%), and beef 37/50 (74%) and the results are presented in table 1.

Table 1: Association of Occurrence of antibiotic residues for different meat types

Meat type	Antibiotic residues		$\chi^2$	P-value
	Positive	Negative		
Chicken	47 (94%)	3 (6%)	7.8055	0.0202
Fish	43 (86%)	7 (14%)		
Beef	37 (74%)	13 (26%)		

Overall, antibiotic residues in fish in these studies were consistently high, regardless of the type and species of the fish. In most fish ponds in Nigeria, antibiotics are directly poured into pond as part of prophylaxis, this could contribute accumulation of antibiotic residues in fish as some of the agents could remain long period unchanged.

In the present study, the occurrence rate of antibiotic residues in beef samples was 74.8% (37/50) (Table 1). Several previous studies showed meat of different types that contained varying degree of antibiotic residues. Zhang *et al.* (2021) analysed antibiotic residues in meat samples from seven cities in Southern Xinjiang. Sixteen out of 26 (61%) antibiotics were detected, with sulfamonomethoxine having the highest concentration. It is important to note that the studies found different concentrations of antibiotics in the samples using different analytic methods; the concentrations pose significant health risks in the long-term effects of exposure. In a study conducted in South Africa on chicken, pork, and beef samples, showed positive test for antibiotic residues, with overall, ELISA analysis showed that 56%, 34%, 18%, and 25.3% of the samples tested positive for ciprofloxacin, streptomycin, sulphanomide and tetracycline residues respectively (Ramatla *et al.*, 2017). The study found high levels of tetracycline residues in muscle, liver, and kidney samples of chicken, pork, and beef in Mafikeng, South Africa.

A recent study conducted in the Far North Region of Cameroon showed that more than 20.30% of cattle slaughtered are contaminated with the residues of two antibiotics named penicillin G (PEN) and oxytetracycline (OTC), which are the mostly used veterinary drugs in the region. This study determined the presence of antibiotic residues, and could be explained on sharing similar geographical border in the study area, and may likely shared unnecessary use of antibiotic in animal husbandry. Various extensive reviews reported similar pattern of presence of antibiotic residues in meats in Africa (Darwish *et al.*, 2013; Mensah *et al.*, 2014; Okocha, *et al.*, 2018; Mohammed *et al.*, 2019; Thi *et al.*, 2020; Tebug *et al.*, 2022)

In a comparative study of presence of antibiotic residues in goat meat and camel meat in total samples of 100, Mohamed *et al.* (2022) found only 13/100 (13%) with 9 (18%, 95% CI: 8.6 - 31.4%) of meat sampled from camel had antibiotic residue, while only 4 (8% 95% CI: 2.2 - 19.2%) of meat from goats contain antibiotic residue. The wide variation of the detection level could be based on the economic importance of the sample source and how frequently are used, hence, camel and the goat may not likely to be exposed to antimicrobial agents at least for prophylaxis purpose. This study is limited to detection of presence of antibiotic residues in beef and was not able to detect the level and types of antibiotics. It is recommended that further study should be conducted in the study area to detect specific types of antibiotics.

## Conclusion

Antibiotic residues in meat is concerning and the results obtained in this study showed the presence of antibiotic residues in meats in Maiduguri. There is need for more studies on type and concentration of antibiotic residues in Maiduguri and the country at large for effective monitoring and to also guide policymaking as part of global One Health concept.

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## Conflict of Interest

The authors have no conflict of interest to declare.

## Authors' Contribution

AM drafted this work, TI, MMI and FAG extensively reviewed the draft, SMI and MSI supervised the work, MAG, FZM and FML participated in the sampling, HB, UAU, LJ and HB were involved in the laboratory work, MMO and JMM proof-read the draft. All authors read and agreed with the final draft.

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