integrity Research Journals

Global Journal of Fisheries Science
Volume 3(3), pages 15-26, August 2021
Article Number: BC903C4A1
ISSN: 2782-750X
https://doi.org/10.31248/GJFS2021.023
https://integrityresjournals.org/journal/GJFS

Full Length Research

Assessment of antimicrobial drugs use and their residue in the farmed fish of Kaduna, Nigeria

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Received 30th April, 2021; Accepted 21st May, 2021

ABSTRACT: The study assessed antimicrobial drug use, pattern, and the occurrence of residues in farmed fish, as well as the level of awareness of drug residues amongst fish farmers in Kaduna State. A total of 300 fish samples from 30 farms in the three senatorial districts of Kaduna State were screened for the presence of antimicrobial drug residues. One hundred questionnaires were administered to fish farmers. Out of the 300 fish samples screened, antimicrobial drug residues were detected in 73.70%. An overall prevalence of 73.70% was obtained, while the prevalence of 63, 74, and 84% was obtained from Kaduna North, Central, and South senatorial districts respectively. From the questionnaire responses, the use of antimicrobial drugs in fish ponds was common (91.86%); with tetracycline (63.41%) being the most frequently used. Self-prescription and administration of drugs by farmers (67.86%) were more in number than those who consult veterinarians for prescription and drug administration. The level of awareness on antimicrobial drug residues in fish and dangers associated with the use and consumption of these residues were very low. Furthermore, the majority of the farmers did not observe antibiotic withdrawal periods before sales of fish to consumers. This study has established a high prevalence of antimicrobial drug residues in farmed fish in the study area. In addition, awareness of the dangers and public health implications of antimicrobial drug residues in fish amongst farmers in Kaduna is low. Therefore, the use of the antimicrobial drug in aquaculture should be regulated and farmers should be encouraged to use immunostimulators or probiotics for disease control and prevention, to safeguard the general populace from the hazards associated with drug residues in fish.

Keywords: Antimicrobial residues; farmed fish, Kaduna, Nigeria.

INTRODUCTION

Aquaculture is currently attracting much attention as an alternative to capture fisheries (Lovell, 1991). A growing amount of fish bought at the marketplace is coming from fish farms. The use of anti-microbial compounds in aquaculture to prevent or treat fish diseases is not unusual and sometimes necessary (Kelly et al., 2020). Following the discovery of the growth-promoting and disease-fighting capabilities of antibiotics, fish farmers, and livestock producers began to use drugs in animal feeds. Antibiotics have been used to treat humans as well as animals such as domestic animals, livestock, and aquatic animals such as fish for many years to prevent or cure infections among the stock population (Horinek, 2009). The major route of transmission of resistant micro-organisms from animals to humans is through the food chain, and micro-organisms also have the potential to be resistant to many types of antibiotics (Akinbowale et al., 2006). Pathogenic bacteria in some species of fish have been linked to bacteria in humans, hence, there should be a concern for the safety of humans (Boinapally and Jiang, 2007).

As in other animal production sectors, antibiotics are used in aquaculture during both production and processing,

mainly to prevent (prophylactic use) and to treat (therapeutic use) bacterial disease contamination (FAO, 1997). Antibiotics have not always been used responsibly in aquaculture, and in many reports, the control of the use of antibiotics has not provided a proper assurance of the prevention of risks to humans (Mensah et al., 2019). Several national governments have already raised the issue of the irresponsible use of antibiotics in all production sectors, with particular concern for the potential risks to public health, and have introduced, changed, or tightened the nation's regulations on the use of antibiotics in general and within the aquaculture sector particularly (FAO, 1997; USFDA, 1997). The unintentional consumption of antibiotics in the food chain is resulting in the development of antibiotic resistance in bacteria pathogenic to humans and this poses a serious risk to human health globally (Angular and Griffin, 2000). Recognition of the risks associated with the direct and indirect effects on human health of both active and passive consumption of antibiotics has led to bans on the use of certain antibiotics in animal food production and the establishment of Maximum Residue Limits (MRLs) for those with known risks (USFDA, 1997; FAO, 1997; He et al., 2016; Mensah et al., 2019).

Growth promotion has been identified as one of the reasons why producers administer antibiotics to fish, therefore, antibiotic residues are expected to be found in fish, including the edible muscle tissue part (FDA, 1998a). Food inspection services need fast, inexpensive, and reliable screening systems for the testing of fish flesh in place of expensive, time-consuming chemical analytical methods for detection of the presence of antimicrobial residues. The main hazards are antibiotic residues and the development of antimicrobial resistance in bacteria that may be transferred to consumers (Stevens et al., 2012; Mensah et al., 2019). Growing concerns over consumer health-related issues associated with the intake of antibiotic residues found in fish and meat products have led to an increased demand for reliable test methods (Stead and Sharman, 2002). Recently, the Premi®Test was released as a new broad-spectrum microbial screening test for the detection of a large number of the most widely used antibacterial substances in fresh meat, meat products, kidney, fish, and eggs (Geijp et al., 1998). Premi®Test is based on the inhibition of the growth of *Geobacillus stearothermophilus*, a bacterium very sensitive to many antibiotics and sulpha compounds (Korsrud et al., 1998). Premi®Test is used to prevent any edible animal product containing antibiotics above the legal limits from entering the food chain. It is a fast, sensitive, reliable, easy-to-use, and cost-effective test covering a broad range of antibiotics (Stead and Sharman, 2002). Where conventional tests require overnight incubation (Romero-González et al., 2007), Premi®Test gives a reliable result in less than four hours. Using this test allows quick decisions about further processing of fish or meat and their products. Premi®Test kit can be used for screening single or large numbers of fish or meat samples (Stead and Sharman, 2002).

The ingestion of antibiotic-contaminated fish has the potential of altering the human gut flora, thus favoring infections of micro-organisms such as Salmonella (Cabello, 2003; 2006). This may increase the risk of toxic and allergic phenomena that may be difficult to diagnose due to the paucity of information on the ingestion of these agents (Sorum, 2006). The excessive use of antibiotics in aquaculture has had negative effects on human and animal health and has also caused damage to the environment (Cabello, 2006). Recent epidemics of bird flu that ravaged the poultry industry discouraged many farmers from poultry keeping and fish production is now becoming popular. This is associated with increased demand for fish and fish products in a country like Nigeria with a rapidly growing population (FAO, 2004). Most part-time fish producers are untrained and do not adopt good management practices and biosecurity measures (FAO/WHO, 2003; Amao et al., 2006). Due to the risks associated with indiscriminate use of antibiotics, it is necessary to research to determine the presence of antibiotic residues in fish from commercial fish farms, and also the level of awareness of the populace on the deleterious effects of antibiotic residues (Olatoye and Basiru, 2013; Belton et al., 2018). Furthermore, there is no government regulation on the use of drugs in fish in Nigeria. There also appears to be no record in the available literature on the extent of use of antibiotics and occurrence of residue in farmed fish in Kaduna State, Nigeria hence the need for this study.

MATERIALS AND METHODS

Study area

Kaduna State is located in the northwestern geographical zone of Nigeria. It lies between latitudes 09° 02'' north and 11° 23'' north and longitude 06° 15'' east and 08° 60'' east and shares boundaries with Katsina, Kano, and Zamfara States to the North, Plateau, and Bauchi states to the East, Nasarawa state, and the Federal Capital Territory to the South and Niger state to the west. The State covers an area of approximately 45,567 km^2, and has an estimated human population of 6,066,562 people according to the 2006 census figures, with three (3) senatorial zones and 23 Local Government Areas (KDSG, 2008). Climatic conditions in Kaduna State are tropical with well-defined wet and dry seasons, the dry windy season, and the rainy (wet) seasons. On average, the State enjoys a rainy season of about five (5) months. There is heavy rainfall in the southern parts of the state like Kafanchan and northern parts like Zaria with an average rainfall of about 1,016 mm (KDSG, 2008) contributes enormously to the economy of Kaduna state with about 80% of the people actively engaged in farming. Another major occupation of the

people is animal rearing and poultry farming; however, aquaculture is emerging. The animals reared include cattle, sheep, goats, and pigs (KDSG 2008).

Sampling design and sample size

A cross sectional descriptive study was conducted. Sample size estimation for the fish was determined using the formula as described by Thrusfield (2005);

$$n = \frac{z^2 pq}{d^2}$$

Where: n = sample size, z = desired confidence 1.96, p=prevalence, q = 1- p and d = allowable error 5%.

Sampling technique

A probability sampling procedure using the convenience sampling technique was employed for the three senatorial zones so that each zone is adequately represented in the sampling. The calculated size was 300 fish samples; however, 314 fish were sampled to avoid sampling errors. Ten (10) fish farms were sampled in each of the senatorial zones while a minimum of ten (10) fish between the ages of 3 to 6 months old (500 g to1 Kg) were collected per farm. Fish (Tilapia and catfish) were sampled from earthen ponds, concrete ponds, and plastic tanks.

Sample collection

Fish samples reared in an intensive system were collected from the ponds by conventional techniques, which is by using a scoop net and transported to the Department of Veterinary Public Health and Preventive Medicine Laboratory of Ahmadu Bello University, Zaria in black 50-liter jerry cans (10 fish per 50-liter jerry can) to reduce stress.

Determination of fish morphometric indices

The weights of the fishes were determined by table scale in grams and kilograms, while the total length (length between the head and tip of tail) was taken by measuring tape. The total length was recorded in centimeters.

Determination of the presence of antibiotic residues in raised fish

Live fish samples were screened for antimicrobial drug residues using a microbial inhibition-based broad-spectrum antimicrobial residues screening test kit (Premi®Test kit, DSM Netherlands). The Premi®Test kit was purchased directly from the manufacturer and stored as directed until use. The principle of the Premi®Test is based on the inhibition of the growth of *Geobacillus stearothermophilus*, a micro-organism very sensitive to many antibiotics and sulphonamide residues. A standardized number of spores are embedded in an agar medium with selected nutrients. When meat fluid is added to the Premi®Test and heated at 64°C, the spores will germinate. The germinated spores will multiply and form an acid when no inhibitory substances are present in sufficient amount (above the detected level), no growth will occur and the color will remain purple. Muscle tissues at the dorsum of the fish close to the gills were collected from the fish samples. The fillet (muscle sample) of each fish was processed separately to reckon with antimicrobial-positive fish. Each sample was tested following the manufacturer's instructions as follows: Muscle tissues were incubated at 64.5°C for 15 min to facilitate the production of liquor. After incubation, liquor was collected by compressing the fish muscle in a juicer. Until testing, liquors were stored at a temperature of -20°C. The Premi®Test consists of an array of tubes containing a test organism immobilized in agar. The tubes were organized in trays of ten by five tubes. One hundred microlitres of the fish fillet liquor were added to each test tube. The test tubes were pre-incubated at ambient temperature for 15 to 20 minutes. Incubation was continued at 64°C for approximately three hours and the color was monitored as follows:

1. A clear color changing from purple to yellow indicated that the antibiotic present is below the Premi®Test detection limits.
2. A purple color indicated the presence of antibiotics at or above the detection limit of the test. The very moment the blank test samples started changing from purple to yellow, all test tubes were read.

Hands were thoroughly washed before starting the test procedures. The required number of ampoules were cut out without damaging the aluminum foil of the ampoules and then the foils were carefully removed. Approximately 2 cm^3 of muscle tissues at the dorsum of the fish close to the gills was cut and a meat press was used to extract approximately 250 ul of meat fluid per sample. A new disposable tip on the syringe for each sample was used.100 ul of the fluid was pipetted onto the agar in the ampoule and then allowed to stand at room temperature for 20 minutes for pre-incubation. The meat juice was flushed away by gently filling and emptying the test ampoules twice with demineralized (demi) water. The ampoules were washed with demineralized water only. The last wash water was carefully removed from the test ampoule. The test ampoules were incubated in the Premi®Test incubator and water bath (64±0.5°C).

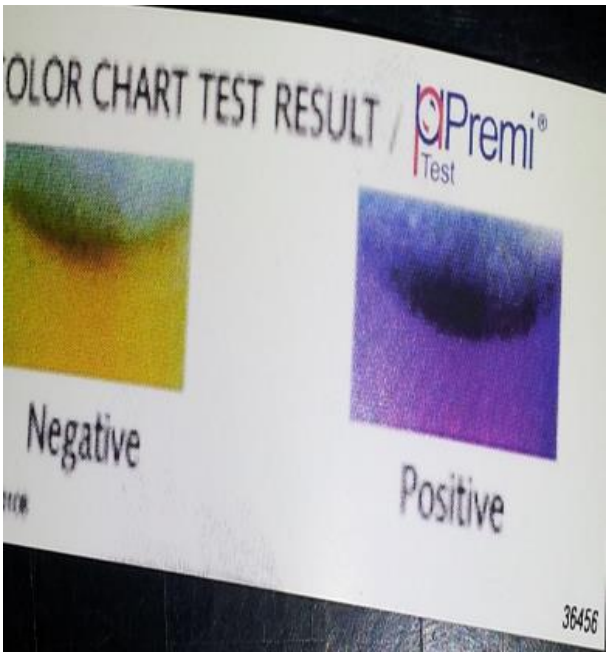

Figure 1. A Premi (R) Test color chart is used for the identification of a positive or negative presence of antimicrobial drug residue in fish.

Determination of the pattern and conditions of antimicrobial drug use and the level of awareness amongst key stakeholders in fish production

The pattern of use of antibiotics in fish farms and the level of public awareness on the deleterious effects of antibiotic residues in fish and fish by-products were studied using a structured questionnaire which was pretested and adjusted to correct limitations identified during pretesting (Figure 1). The questionnaire gathered information on demographic characteristics of respondents involved in fish production including fish farmers, veterinarians, veterinary assistants, and other respondents. Information on fish production and management practices, fish health, and drug usage were obtained. Additional questions were also asked, when necessary, to shed more light on some issues raised during questionnaire administration.

Data analyses

The data generated were analyzed by descriptive statistics and cross-tabulation using SPSS (version 17). Chi-square test was used to determine the association between categorical variables at a significant level of $p<0.05$.

RESULTS

Prevalence of antimicrobial drug residues in fishes

Out of the 300 fishes tested, the antibiotic residue was detected in 221 (73.70%) with an overall prevalence of 63, 74, and 84% in Kaduna North, Central, and South senatorial districts respectively. There was also a statistically significant difference ($\chi 2 = 11.38$, $df = 2$, $p<0.05$) between senatorial district and detection of antibiotic residue in the fishes (Table 1). All the fishes were table size ready for market with a mean weight and total length of 0.66±0.01 kg (range 0.2 to 1.2 kg) and 35.06±0.34 cm (range 18 to 50 cm) respectively.

The pattern of antimicrobial drug use in fish farms in Kaduna State

Out of the 100 questionnaires distributed to investigate the pattern of use of antimicrobial drugs in fish farms in the study area, 88 of the questionnaires were retrieved and evaluated, the remaining 12 could not be retrieved due to logistic problems.

Demographic characteristics

The demographic characteristics of the fish farmers indicated that the majority (81.8%) were between the ages of 20 to 59, while 5.70 and 12.5% respectively were less than 20 years and 60 years (Table 2). About 66.67% of the farmers were males and 33.33% were females. Regarding the highest level of education attained by the farmers, the majority (68.60%) had tertiary education, 27.90% had secondary education, and 2.30% had primary education while 1.16% had no formal western education. The primary occupation of the respondents included; civil servants (27.27%), farmers (25.00%), businessmen/women (35.23%) and artisans 12.50% (Table 2).

Farm management practices

An intensive system of farming was practiced by 89.53% of the farmers while 8.14 and 2.33% practiced semi-intensive and extensive farming systems respectively. Commercial purpose was the reason for fish farming among 90.70% of the fish farmers, subsistence by 4.65%, and both commercial and subsistence by 2.31% (Table 3). Catfish (*Clarias gariepinus*) was the major (72, 84.70%) fish species kept by the farmers. Tilapia was kept by only by a small proportion (4, 4.71%) while 9 (10.59%) kept both catfish and Tilapia. Forty-nine (56.98%) kept only fish in the farms, 27 (3.40%) kept both fish and poultry, while 10 (11.63%) kept fish and other livestock. A major percentage of the farmers; 76.74 and 67.01% respectively had other fish farms surrounding theirs and do not allow other animals access into their fish ponds (Table 3). Seventy-one (88.75%) of the farmers had between 1 to 10 fish ponds in their farms with 55 (63.95%) having a fish-holding capacity of between 500 to 2000 fishes. Concrete, earthen and plastic ponds were commonly used by 28

Table 1. Prevalence of antimicrobial drug residues among fish samples from the three senatorial districts of Kaduna State, Nigeria.

Senatorial district	Total number of fish samples	Samples positive for drug residue (%)
North	100	63 (63)
Central	100	74 (74)
South	100	84 (84)
Total	300	221 (73.70)

$\chi^2 = 11.38$, df = 2, $p<0.05$.

Table 2. Demographic characteristics of the stakeholders in the fish industry in Kaduna State, Nigeria.

Demographic variables	Frequency (%)
Age	
< 20	5 (5.70)
20-59	72 (81.80)
60 and above	11 (12.50)
Sex	
Males	58 (66.67)
Females	29 (33.33)
Educational status	
Primary Education	2 (2.30)
Secondary Education	24 (27.90)
Tertiary Education	59 (68.60)
None	1 (1.16)
Primary occupation	
Civil Servant	24 (27.27)
Farmer	22 (25.00)
Business man/woman	31 (35.23)
Artisan	11 (12.50)

(32.56%), 13 (15.12%), and 20 (23.26%) respectively of the farmers for rearing fishes while 25 (29.07%) use a combination of the three different types of ponds (Table 3). Antimicrobial drug residues were also found in the fish water and feed in almost equal percentage for good and low quality water, but in higher percentage in pelleted feed compared to extruded feed (Table 4). This implies that all the channels of fish production have antimicrobial drug residues .

Antimicrobial drug use by farmers in fish production

Fifty-one (59.30%) of the fish farmers reported they had 2 to 5 outbreaks of disease in their farms in the last 6 months, while 7 (8.14%) had not encountered any disease outbreak in the last 6 months. The farmers (7, 8.33 %) reported that when they encounter such episodes of

Table 3. Farm management practices among fish farmers in Kaduna State, Nigeria.

Variables	Frequency (%)
Farming system	
Intensive	77 (89.53)
Semi-intensive	7 (8.14)
Extensive	2 (2.33)
Purpose of fish farming	
Commercial	78 (90.70)
Subsistence	4 (4.65)
Both commercial and subsistence	2 (2.31)
Fish species kept	
Tilapia	4 (4.71)
Catfish	72 (84.70)
Mixed	9 (10.59)
Type of livestock kept	
Fish only	49 (56.98)
Fish and poultry	27 (31.40)
Fish and other livestock	10 (11.63)
Presence of other fish farms around the farm	
Yes	66 (76.74)
No	20 (23.26)
Access by other animals into the fish pond	
Yes	28 (32.94)
No	57 (67.01)
No of ponds on the farm	
1-10	71 (88.75)
11-20	6 (7.50)
>20	3 (3.75)
Pond type	
Concrete	28 (32.56)
Earthen	13 (15.12)
Plastic/fiber tanks	20 (23.26)
Combination of the above	25 (29.07)
Pond holding capacity	
< 500	20 (23.26)
500-2000	55 (63.95)
2001-4000	9 (10.47)
>4000	2 (2.33)
Marking outlets	
Hotels and restaurants	4 (4.71)
Markets	35 (41.18)
Households	7 (8.21)
Combination of the above	39 (45.88)

Table 4. Relationship between farm management practices and detection of antimicrobial drug residues in farmed fish from Kaduna State, Nigeria.

Management practices	No. of positive farms (%)
Water quantity	
Adequate	22 (81.48)
Low	1 (3.70)
Moderate	4 (14.81)
Water quality	
Low	13 (48.12)
Good	14 (51.85)
Feed type	
Pelleted	17 (62.96)
Extruded	9 (33.33)
Pelleted and Extruded	1 (3.70)
Feeding characteristic	
Overfeeding	12 (44.44)
Normal	14 (51.85)
Underfeeding	1 (3.70)
Stocking density	
High	16 (59.26)
Adequate	8 (29.63)
Low	1 (3.70)
Overstocking	2 (7.41)

disease, it usually occurs frequently and 60.27% (44) said it affects less than half of the fishes. Seventy-nine (91.86%) of the farmers have used antibiotics/drugs in the fish ponds for disease treatment (86.75%), growth promotion (3.61%), prophylaxis (4.82%), and another 4.82% for the triad of treatment, prophylaxis, and growth promotion (Table 5). Tetracycline was the most commonly (63.41%) used antibiotic in the fish ponds by the farmers and they applied antibiotics/drugs once weekly (7.32%), twice weekly (14.63%), once monthly (43.90%), yearly (15.85%) and occasionally (18.29%) into the fish ponds (Table 5). From the results obtained, veterinarians, animal scientists, livestock superintendents, fellow farmers and the farmers were responsible for recommending (11.90, 16.67, 2.38, 1.19 and 67.86 %, respectively) and administering (27, 39.29, 8.33, 13.10 and 11.90%, respectively) the antimicrobial drugs (Table 6). Water was reported as the major (66.00 %) route of administering these antibiotics/drugs, followed by feed (14.00%) and a combination of water and feed by 20% of the farmers (Figure 2).

Awareness of antimicrobial drug residue in food and its potential dangers

The majority (67.00%) of the fish farmers were not aware

Table 5. The pattern of disease outbreak and antimicrobial drug use in fish farms in Kaduna State, Nigeria.

Variables	Frequency (%)
Disease episode in the farm in the last 6 months	
No episode	28 (32.56)
2-5	51 (59.30)
Never	7 (8.14)
Frequency of disease outbreak	
Frequently	7 (8.33)
Occasionally	53 (63.10)
Rarely	16 (19.05)
Never	8 (9.52)
The population of fish affected	
All	3 (4.11)
Majority	8 (1.10)
Half	18 (24.66)
Less than half	44 (60.27)
Ever used antibiotic/drug in fish ponds	
Yes	79 (91.86)
No	7 (8.14)
Type of antibiotic/drug use	
Tetracycline	52 (86.75)
Sulphonamides	2 (2.44)
Other (Fish salt, perflox)	28 (34.15)
Purpose of antibiotic/drug use	
Disease treatment	72 (86.75)
Promote growth	3 (3.61)
Prophylaxis	4 (4.82)
All of the above	4 (4.82)
Frequency of antibiotic/drug use	
Once weekly	6 (7.32)
Twice weekly	12 (14.63)
Once a month	36 (43.90)
Yearly	13 (15.85)
Occasionally	15 (18.29)

nor have they heard of antibiotic/drug residue in food (Figure 3). Twenty-seven percent of the respondents heard of drug residue from veterinarians, friends, and print media, 13% got this information from the school environment while 3% each got this information from fellow farmers and the electronic media (Figure 4). There was no statistically significant association ($p>0.05$) between sex of the respondents ($\chi 2 = 0.563$, $p = 0.755$), primary occupation of the respondents (Fisher's exact test $p = 0.649$) and awareness of drug residue (Table 7). Sixty-four

Table 6. Personnel is responsible for recommending and administering antimicrobial drugs for fish farmers in Kaduna State, Nigeria.

Variables	Frequency (%)
Who recommends the antibiotic/drug to be used?	
Veterinarian	23 (27.00)
Animal scientist	33 (39.29)
Livestock superintendent	7 (8.33)
Fellow fish farmer	11 (13.10)
Self	10 (11.90)
Who administers the antibiotic/drug?	
Veterinarian	10 (11.90)
Animal scientist	14 (16.67)
Livestock superintendent	2 (2.38)
Fellow fish farmer	1 (1.19)
Self	57 (67.86)

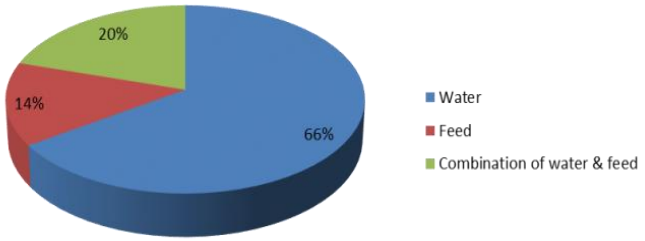

Figure 2. Common routes of antimicrobial drug administration by farmers in fish farms in Kaduna State, Nigeria.

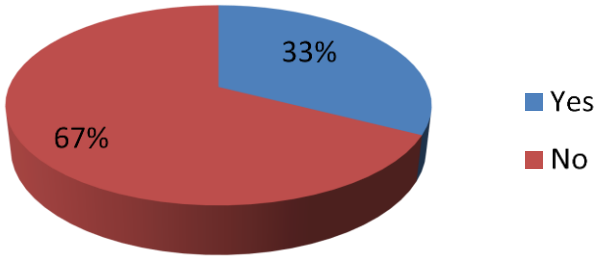

Figure 3. Awareness of antimicrobial drug residues among fish farmers in Kaduna State, Nigeria.

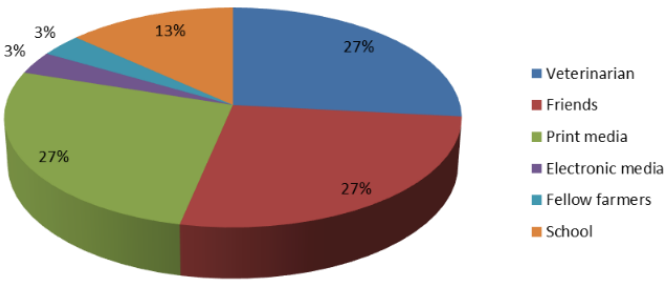

Figure 4. Respondents' source of awareness on antimicrobial drug residues in fish farms in Kaduna State, Nigeria.

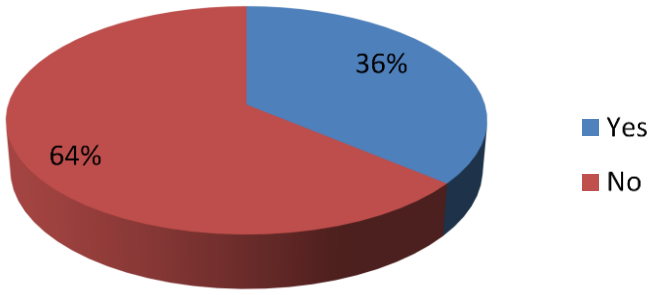

Figure 5. Awareness of the dangers associated with antimicrobial drug residues among fish farmers in Kaduna State, Nigeria.

percent of the respondents were not also aware of the dangers associated with antimicrobial residue while 36% were aware (Figure 5). Sixteen (48.48%), 8 (24.24%), and 9 (27.27%) of the respondents that knew the dangers reported drug resistance and hypersensitivity and a combination of drug resistance and hypersensitivity respectively as the dangers associated with consumption of antibiotics residues in food (Table 8). The majority (58.62%) of the respondents were not sure if antibiotics/ drug residues can be ingested with fish, 31 (35.63%) were sure while 5 (5.75%) said it can not be consumed with fish. Twenty-six (30.23%), 7 (8.14%) and 53 (61.63%) of the respondents think, do not think, and not sure respectively that drug residue in fish is a serious health problem (Table 8). Also, 28 (35%), 19 (23.75%), 30 (37.50%), and 3 (3.75%) of the respondents respectively sell out their fishes for consumption during treatment with antibiotics, a few days after treatment with antibiotics, more than one week after treatment with antibiotics and a combination of the above (Table 8).

DISCUSSION

This study was aimed at the assessment of antimicrobial drug use and the occurrence of residues in farmed fish in Kaduna State, Nigeria. In the present study, the overall prevalence of antimicrobial drug residues from farmed fish in Kaduna State was 73.70% from the 300 fish samples screened, with the prevalence of 63, 74, and 84 from Kaduna North, Kaduna Central, and Kaduna South respectively. This shows that antibiotic residues are present in farmed fish in Kaduna State. The overall prevalence obtained in this study is higher than the result (52.5%) obtained by Olatoye and Basiru (2013) in Ibadan. Because they screened only catfish (*Clarias gariepinus*) from farms and restaurants. In this present study, both catfish and Tilapia were screened and all were from farms. In both studies, Premi®Test was used for the screening, but Olatoye and Basiru (2013) used the high-performance liquid chromatography (HPLC) to specifically screen for tetracycline. The presence of antimicrobial residues in farmed fish in the present study concur with other reports

Table 7. Relationship between awareness of antimicrobial drug residues among fish farmers and demographic characteristics.

Demographic variables	Aware	Not aware	x^2-value	P-value
Sex				
Male	18	39	0.563	0.755
Female	10	19		
Age				
< 20	0	5		
20-59	25	46		
60 and above	3	7		
Level of education				
None	0	1		
Primary	1	1		
Secondary	2	21		
Tertiary	25	33		
Primary occupation				
Civil servant	9	15		
Farmer	8	14		0.649*
Businessman/woman	7	22		
Artisan	4	7		

*Fishers Exact test.

Table 8. Beliefs and practices associated with antimicrobial drug residues among fish farmers in Kaduna State, Nigeria.

Variables	Frequency (%)
Dangers associated with consumption of antibiotic residue in food	
Drug resistance	16 (48.48)
Hypersensitivity reactions	8 (24.24)
Combination of the above	9 (27.27)
Can one ingest antibiotics/drugs via the consumption of fishes?	
Yes	31 (35.63)
No	5 (5.75)
Not sure	51 (58.62)
Do you think antibiotic/drug residue in fish is a serious health problem?	
Yes	26 (30.23)
No	7 (8.14)
Not sure	53 (61.63)
How long before you sell you fishes after treatment with an antibiotic?	
During treatment	28 (35.00)
Few days after treatment	19 (23.75)
More than one week after treatment	30 (37.50)
Combination of the above	3 (3.75)

of antimicrobial residues in food animal production in Nigeria (Dipeolu and Alonge, 2002; Kabir et al., 2004; Fagbamila et al., 2010; Ezenduka et al., 2011; Olatoye and Saraye, 2012) and other countries (FDA, 1998b; FAO , 2002; Okerman et al., 2004; Cabello, 2004; Bagumire et al., 2010; Chafer-Pericas et al., 2010a, b; Mensah et al.,

2019). It is assumed that the high prevalence of antimicrobial drug residues observed in this study may be due to the indiscriminate use of these drugs by fish farmers (Choo, 1995), and also the non-observance of drug withdrawal period after treatment and before the fish are sold to consumers as observed in the questionnaire responses of the farmers in the present study and reported by Navarrete (2008). Besides, a low prevalence of antimicrobial drug residue was obtained from Kaduna North as compared to that obtained from other senatorial districts. It is presumed the presence of a Veterinary Teaching Hospital and a Veterinary Clinic in Kaduna North may have influenced the use of antimicrobial drugs by farmers in this area. The farmers in this zone may have routinely sought professional advice and have access to veterinary services. Furthermore, Kaduna South in the present study had the highest prevalence (84%) of antimicrobial drug residues in farmed fish. The high prevalence recorded in this senatorial zone could be attributed to the non-availability of potable water for their ponds and the fewer number of veterinarians that may have rendered veterinary service to their farms. Thus, farmers resorted to the use of readily available contaminated water. Consequently, this may lead to increased disease incidence in ponds (Oladosu and Ajasin, 2012) due to the increased occurrence of pathogenic organisms (Dietz et al., 2005). Hence, fish farmers are obliged to use antimicrobials frequently. Further still, the low number of veterinarians in Kaduna South may have limited the access of fish farmers to professional veterinary services, which culminated in farm management practices that are not in tandem with recommended standard practices in the use of antimicrobials. According to Hernandez (2005) and Cabello (2006), several classes of antimicrobial drugs are commonly used in large quantities in fish farming especially in developing countries including Nigeria where the use of these drugs is not regulated. Similarly, when antimicrobials are mixed with fish feed as growth promoters (Svobodova et al., 2006; Kan and Meijer, 2007), residues may be deposited in the meat, and consumers may inadvertently consume fish containing these residues, which may lead to changes in their microbial flora (Uma and Rebecca, 2018), thus altering the intestinal ecology and favoring the emergence of resistant microflora and drug allergies such as hypersensitivity reactions and other human hazards (Berends et al., 2001).

In the present study, 66.7% of stakeholders interviewed were males while 33.33% were female. The mean age of fish farmers in this study concurs with that reported by Olatoye and Basiru (2013) in Ibadan. However, the average years of fish production experience differ in both studies. The numbers of fish farmers (91.86%) who engaged in routine use of antimicrobial drugs to control and prevent diseases in their ponds were slightly higher than 90% reported by Olatoye and Basiru (2013) in Ibadan. Aquaculture is a rapidly growing business in Nigeria and Kaduna State in particular (Omole et al., 2006; Amao et al., 2006; Kehinde et al., 2009; Oladosu and Ajasin, 2012). Fish farming is attracting the attention of a wide range of people with different educational qualifications and occupations. Therefore, the increasing growth of fish farming in Nigeria and Kaduna State could lead to pollution of the aquatic environment with huge amounts of antimicrobial drug and consequently, their residues in fish and fish products (Hirsch et al., 1999). The most frequently used antimicrobial agent found in this study was oxytetracycline, which is in agreement with previous reports (Okerman et al., 2004; Mo et al., 2017; Okocha et al., 2018; Kelly et al., 2020). Oxytetracycline being a broad spectrum and relatively cheap antibiotic is widely used by farmers in both fish and livestock production (Olatoye and Ehinmowo, 2010). Thus, it may be right to presume that tetracycline is the most misused antimicrobial drug in aquaculture. The number of fish farmers (67.80%) practicing self-administration of antimicrobials in Kaduna State is higher than those reported in Ibadan (Olatoye and Basiru, 2013). Additionally, the observance of the drug withdrawal period by farmers was low in Kaduna State. Similarly, awareness of the potential hazards of antimicrobial drug residues in humans was low. The majority (67%) of farmers in the present study were ignorant of the presence and public health implication of antimicrobial drug residues in fish (Bagumire et al., 2010; Henriksson et al., 2017). Despite their high educational attainment (68.60 %), most fish farmers in Kaduna State, surprisingly, still engaged in self-medication and misuse of antimicrobial drugs as a routine practice without proper diagnosis. Furthermore, they did not observe the withdrawal period after treatment. This may be another reason that may be attributed to the high prevalence obtained in this study. Also, farm management practices as observed in this study can influence the presence of residues in fish and the consequence of resistant pathogens.

Admittedly, the high prevalence of antimicrobial drug residues obtained in this study using Premi®Test as the screening method for antimicrobials in fish confirms the suitability and further validate the sensitivity of the test as argued by Okerman et al. (2004) and Olatoye and Basiru (2013). Premi®Test has been reported to be sensitive for detection of several groups of antimicrobial agents (Olatoye and Basiru, 2013; Chereau et al., 2017) . Further still, Okerman et al. (2004), Han et al. (2017), and Miller and Harbottle (2018) argue that the efficient control of drug residues in foods of animal origin requires good screening tests, which must be cheap, easy to perform, allow for simultaneous analyses of large numbers of samples and give rapid results. Unfortunately, there is no available information on the survey, monitoring, quantitative analyses of antimicrobial drug residues in farmed fish in Kaduna State. Admittedly, the poor enforcement of regulations on veterinary drug handling and use in Nigeria could perhaps account for the high level of antimicrobial

drug residues obtained in farmed fish in Kaduna State. Therefore, the use of antimicrobial drugs in aquaculture should be a great concern as it may lead to antimicrobial drug-resistant pathogens and residues in the food chain. The results obtained in the present study have confirmed that there may be a misuse of antimicrobial drugs in farmed fish in Kaduna, Nigeria.

Conclusions

This study has established the following: Antimicrobial drug residues are present in farmed fish in Kaduna State, Nigeria with an overall prevalence of 73.70%. Kaduna South had the highest prevalence (84%) of antimicrobial drug residues, followed by Kaduna Central with 74% and Kaduna North with 63%. Fish farmers in Kaduna State are engaged in uncontrolled and unsupervised administration of antimicrobial drugs in aquaculture production (91.86%). Awareness of the dangers and public health implications of antimicrobial drug residues in fish is low amongst fish farmers in Kaduna State (63%), this may be a contributory factor for the high level of antimicrobial drug residue. Proper use of the antimicrobial drug, vaccination, the use of immune stimulants and probiotics for disease control and prevention in aquaculture should be encouraged. Fish farmers should be educated on the need to always observe withdrawal periods after treatment and before the sale of treated fish to consumers. Government should enforce strict regulation on the use of veterinary drugs in fish farming; educate fish farmers on the dangers inherent in antimicrobial drug residues in fish to safeguard the general populace from hazards associated with the consumption of antimicrobial drug residues. Routine antimicrobial drug residue screening of fish meant for human consumption is advocated. Use of antimicrobial drugs for the purposes such as growth enhancement, preservation, etc. other than treatment should be discouraged. Further studies to determine the specific antimicrobial drug residues in farmed fish in Kaduna State are required.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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