# SCREENING OF ANTIMICROBIAL RESIDUE IN COMMERCIAL EGGS IN MAIDUGURI METROPOLIS, BORNO STATE

#### 4 ABSTRACT

5 The objectives of the study were to screen for antimicrobial residue in table eggs in Maiduguri 6 metropolis. Multistage sampling technique was used based on the 4 major district of Maiduguri 7 Metropolis Viz; Bolori, Gwange, Kyarimi park and Shehuri North. Four hundred commercial egg 8 samples were collected for the study. One hundred and sixteen table eggs were sampled from 35 9 randomly selected poultry layer farms and 284 were obtained from 37 randomly selected egg 10 commercial retail outlets. The antimicrobial screening of eggs was carried out using the disc diffusion 11 method where Bacillus cereus ATCC 14579 from spectra medics' laboratory in Ogun state was used 12 as the test organism. One hundred and sixteen (116) table eggs collected from farms across the study 13 district, 36 (31 %) were each from Bolori and Gwange, 39 (33.6 %) from Kyarimi Park and 5 (4.3 %) 14 from Shehuri North. A total of 49 positive samples were obtained which include 17 (47.2 %), 21 (58.3 15 %), 10 (25.6 %) and 1 (20 %) from Bolori, Gwange, Kyarimi Park and Shehuri North respectively 16 (Figure 2). There was no significant difference (P=0.095) among the clusters. Out of the 284 egg 17 samples collected from the retail outlet, 201 (70.1 %) samples were from Jos and 83 (29.2 %) from 18 Ibadan. A total of 100 (35.2 %) samples were positives for antimicrobial screening which comprises 19 of 71 (35.32 %) and 29 (34.94 %) from Jos and Ibadan respectively. With no significant difference 20 between the two sources (P=0.902). From this study, it was concluded that: There is small flock size 21 (backyard) farm in Maiduguri with 94.3 % of the farmers holding equal or less than 500 birds in their 22 farms. Antimicrobial residue detected in the study area is alarming.

23 Keywords: Table egg, Bacillus cereus, Maiduguri Metropolis, Antimicrobial residue

# 24 Introduction

25 Egg contains carbohydrate, protein and other essential substances required for human existence (Braun, 2000). The low caloric value, edibility and nutrient content makes egg 26 27 significant foodstuff for many dietary regimes (Kenner et al., 2006). The hen's egg is a 'selfcontained unit for starting a new life (FAO, 2013). The egg is a major food source, providing 28 good quality balanced nutrient to billions of people throughout the world, the world's total 29 30 hen production in 2011 was 70.5 million tonnes, which is 8 million tonnes more than beef production for the same year (FAO, 2013). Poultry is an essential component of the Nigerian 31 32 economy, providing income for peasant farmers and a good source of high-quality protein for the ever growing population of Nigeria (Agbaje et al., 2010). In livestock production, poultry 33 occupies a prominent position in the provision of animal protein and this account for about 34 35 25% of local meat production in Nigeria (Agbaje et al., 2010). The annual production 36 capacity of commercial eggs in Nigeria is estimated at 8, 216, 208, 000 eggs equivalent to 37 273, 873, 600 crates of eggs (FAO, 2008).

38 Antibiotic usage has facilitated the efficient production of poultry, allowing the consumer to purchase at a reasonable cost, high-quality meat and eggs as well as reduce the impact of 39 40 disease outbreaks (Donogue, 2003; karmi, 2014). They are used by the poultry industry to enhance growth, feed efficiency and reduce bacterial disease (Donoghue, 2003). In layer 41 hens, antimicrobials are only used to treat and prevent bacterial infections. Some of the 42 43 antimicrobial classes used in treating layers include aminoglycosides, tetracyclines, betalactams, quinolones, macrolides, polypeptides, amphenicols and sulphonamides (Stolker and 44 Brinkman, 2005). Through the years the issue of antibiotic residue from farm animals and 45 46 their effect on human health has been a significant concern (Bahry *et al.*, 2013).

The consequences of the substantial use of antimicrobials in laying hens is residue
accumulation in egg (Sirdar *et al.*, 2012). Very few antibiotics are approved for use in laying

hens (Castanon, 2007). In Maiduguri, the study area, antibiotics are freely marketed without
veterinary prescription (Geidam *et al.*, 2012), and despite a report of misuse of antibiotics,
there is paucity of information regarding the level of antibiotic residue in commercial eggs
meant for human consumption in the Maiduguri Metropolis.

#### 53 MATERIAL AND METHOD

#### 54 Study area

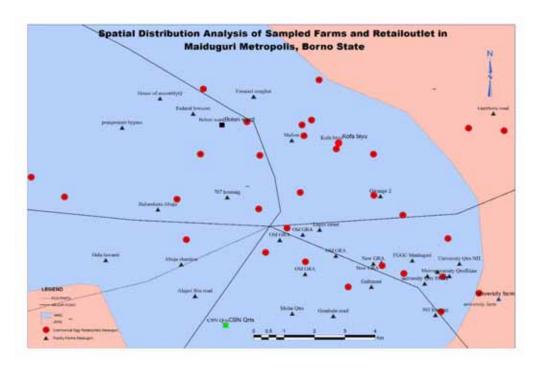
55 Maiduguri Metropolis, a major city in the Northeastern part of Nigeria, is located between 56 latitudes 11.04'N and 11.44'N; and between longitudes 13.04'E and 13.44'E. It covers a total land area of 543 km<sup>2</sup>, which makes it the largest city in the Northeastern region of Nigeria 57 (Daura, 2002; Jimme et al., 2016). Maiduguri city now extends to four Local Government 58 Areas: Maiduguri Metropolitan, Jere, Konduga and to a smaller extent part of Mafa local 59 60 government areas (Daura et al., 2001). The climate of Maiduguri is characterized by a long dry season with high evaporation rate from October to May and a short Wet season for the 61 62 remaining part of the year (Jimme et al., 2016). There are four identified seasons in the area 63 which include the Rainy Season, (June to September) Harvest Season (September to 64 November), Harmattan or Cool Season (December to February) and Hot Season (March to May) (Waziri, 2009). It has a population estimated at 1.275 million people according to the 65 66 2006 census (NPC, 2008). With an annual growth rate of about 3.5% and a density of 1145 67 persons per square km which makes it the most densely populated city in North Eastern 68 Nigeria (Waziri, 2009; Jimme *et al.*, 2016). Crop production and livestock farming are the 69 predominant occupation of the people in the study area (Tijjani et al., 2012). Poultry layer production is a profitable business in Maiduguri Metropolis (Tijjani et al., 2012). 70

# 71 Study design

# 72 Sampling technique

73 Multi stage sampling method was used for sample collection.

74 Maiduguri metropolis was divided into 4 major district by Borno state water board namely 75 Bolori, Gwange, Kyarimi Park, and Shehuri North. In this study, these areas were taken as 76 the primary sampling units. Laying poultry farms and egg retail outlets located in each of the 77 primary sampling units above were taken as secondary sampling units. Fifty per cent of farms 78 and 10 % of egg retail outlets were randomly selected. One egg was collected from 50 laying 79 hens in each selected laying farm and 1 egg out of a crate in the retail outlets was taken as 80 tertiary sampling unit. Geographical coordinates of the sampled areas were taken and 81 recorded. A spatial distribution analysis of the layer farm sampled were constructed (Figure 82 1).



84 **FIGURE : 1** 

#### 85 Sample size determination

- 86 The determination of sample size for table eggs collection was based on the formula given by
- 87 Thrusfield (1995) for simple random sampling method.

$$n = \frac{z^2 p q}{d^2}$$

88 Where:

n = sample size

- 90 z=desired confidence 1.96
- 91 p=prevalence= 3.6 % by Fagbamila et al., (2010) in Jos plateau state.

92 q= 1-p

93 d=allowable error 5 %

Thus a total sample size of 180 table eggs was determined and was rounded up to 200 samples for convenience. The sample size was inflated 2 times to increase precision (2 x 200) reaching 400 table eggs to be sampled for the study (Thrusfield, 2005).

#### 97 Sample collection

A total of four hundred table egg samples were collected, out of which 116 samples were 98 from 35 layer poultry farms and 284 table eggs were from 36 retail outlets in the four major 99 100 areas of Maiduguri Metropolis. Fifty percent of layer farms and 10 % retail outlets were 101 selected from each cluster. One table egg was collected in each fifty laying hens from the 102 selected farms and the sampling covered a period of 3 weeks. One table egg was collected 103 from each crate containing 30 eggs from selected retail outlets and the sampling covered a 104 period of 2 weeks. The Table eggs collected were arranged in a clean crate, labeled and 105 transported to the veterinary medicine laboratory immediately for processing.

# 106 Sample processing

107 The antimicrobial screening of eggs was carried out using the disc diffusion method where 108 Bacillus cereus ATCC 14579 from spectra medics' laboratory in Ogun state was used as the 109 test organism. An 18 hour culture of the test organism in 10 ml nutrient broth (Oxoid Basingstoke, Hampshire, UK) was used to inoculate Mueller Hinton agar plates. The egg 110 111 surface was thoroughly cleansed using sterile cotton wool soaked in 70% alcohol. Sterile 112 forceps were used to puncture the egg at the tip to create a small opening from where the yolk 113 were carefully drained out into a sterile beaker and mixed with Phosphate Buffer Saline pH 114 7.4 and then thoroughly homogenize, then 10 milliliter was transfer to a clean sterile test tube 115 and was centrifuge for 10 minutes at 4000 x g. Two milliliter of the supernatant was 116 transferred to sterile petri dish (Fagbamila et al., 2010; Kabir et al., 2004).

# 117 **Qualitative screening**

Using a clean sterile forceps Whatman<sup>®</sup> filter paper disc 0.6 cm in diameter was dipped into 2 118 mm of the egg supernatant in the Petri dish, until it is soaked and then were exposed to 119 temperature of 80°C for 10 minutes to in activate inhibitory substance and placed gently on 120 121 the Mueller Hinton agar plate that has already been inoculated with the test organism according to the method of Shahid et al., (2007). This was then incubated at 37°C for 24 122 123 hours after which the plates were viewed for the presence or absence of zones of inhibition of 124 the test organisms around discs. Any disc with a zone of inhibition greater than 1 mm around 125 the disc was considered positive (Kabir et al., 2004).

# 126 Data analyses

127 The data was compiled and analysed with Statistical Package (SPSS statistical package 128 version 21). Chi-square was used to determine association between variables at significant 129 level of P < 0.05.

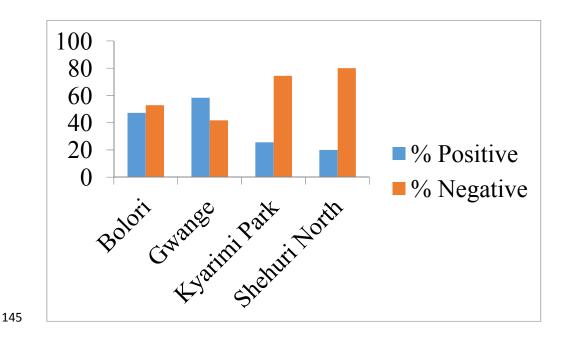
130 **Results** 

131 One hundred and sixteen (116) table eggs collected from farms across the study area, 36 (31 132 %) were each samples from Bolori and Gwange farms, 39 (33.6 %) from Kyarimi Park farms 133 and 5 (4.3 %) from Shehuri North farms. A total of 49 positive samples were obtained which include 17 (47.2 %), 21 (58.3 %), 10 (25.6 %) and 1 (20 %) from Bolori, Gwange, Kyarimi 134 135 Park and Shehuri North respectively (Figure 2). There was no significant difference 136 (P=0.095) among the clusters. Out of the 284 egg samples collected from the retail outlet, 137 201 (70.1 %) samples were from Jos and 83 (29.2 %) from Ibadan. A total of 100 (35.2 %) 138 samples were positives for antimicrobial screening which comprises of 71 (35.32 %) and 29 (34.94 %) from Jos and Ibadan, respectively (Figure 3) with no significant difference of 139 140 residue of antimicrobials between the two sources (P=0.902).

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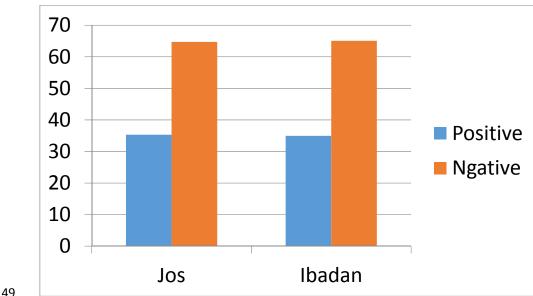
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# 147 Figure 2: Result of qualitative screening for antibiotic residues in table eggs from



148 Poultry Layer farms in Maiduguri Metropolis, Nigeria

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Figure 3: Result of qualitative screening for antibiotics residues from retail outlet in
 Maiduguri Metropolis, Nigeria

### 153 Discussion

154 Occurrence of antibiotics residue in laying hens may be due to failure to observe withdrawal period, extra label dosage, contamination of animal feed with excreta of treated animal or the 155 156 use of unlicensed antibiotics. All the commercial egg samples used for the study were obtained from layer farms and retail outlets. The majority of the retailers source their eggs 157 158 from Jos and Ibadan in order to compensate for the short fall from local production in the 159 study area. Short fall is connected with low flock size in the study area. The finding of this study is similar to that of Tijjani et al., (2012) who reported small flock poultry layer farming 160 161 in Maiduguri Metropolis. Antibiotic residues were detected in 49 (42.2 %) of the eggs 162 samples collected from farms with lower percentage 100 (35.2 %) in egg samples collected 163 from retail outlets. The lower percentages in retail outlets might not be unconnected with the storage or variation of antibiotic use by different farms were the eggs were sourced. This is in 164 165 tandem with observation of Ezenduka et al., (2011) in Enugu who reported 36 % positive in 166 eggs sampled from farms and 30 % in retail outlets and also El-Nasri et al., (2012) in Sudan 167 reported 55.4 % antibiotic residue in eggs collected from farms and 43.2 % in retail outlets. 168 Islam et al., (2016) in Bangladesh reported higher percentage (60 %) of antibiotic residue in 169 table eggs. The research of Kabir et al., (2004), Fagbamila et al., (2010) and Omeiza and Nafarnda, (2015) reported a lower percentage of 0.5 %, 3.6 % and 18.5 % antibiotic residue 170 171 in table eggs respectively. The reason might be due to variation in awareness of biosecurity, 172 antibiotic residue in table eggs and public health effect of antibiotic residue.

173 Conclusion

From this study, it was concluded that: Most of the poultry layer farmers have small flock size (backyard) poultry farm in Maiduguri Metropolis with 94.3 % of the farmers holding equal or less than 500 birds in their farms. Percentage of antibiotic residue detected were 42.2

177 % and 35.2 % in commercial egg samples collected from layer farms and retail outlets178 respectively.

# 179 **Recommendation**

Farmer education on the use of antibiotics and its public health implication. Antibiotics being a prescription drug should not be freely sold to farmers over the counter. More research using sensitive techniques should be carried out to quantify the residue levels of individual prohibited for use in food-producing animals. Antibiotics Legislation regarding the use of prohibited antibiotics on food animals by National Agency for Food and Drug Administration and Control.

#### 186 Ethical Disclaimer:

187 As per international standard or university standard written ethical approval has been188 collected and preserved by the author(s).

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