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

ABSTRACT

The objectives of the study was to screen for antimicrobial residue in table eggs in Maiduguri metropolis. Multistage sampling technique was use based on the 4 major district of Maiduguri Metropolis Viz; Bolori, Gwange, Kyarimi park and Shehuri North. Four hundred commercial egg samples were collected for the study. One hundred and sixteen table eggs were sampled from 35 randomly selected poultry layer farms and 284 were collected from 37 randomly selected egg commercial retail outlets. The antimicrobial screening of eggs was carried out using the disc diffusion method where *Bacillus cereus* ATCC 14579 from spectra medics' laboratory in Ogun state was used as the test organism. One hundred and sixteen (116) table eggs collected from farms across the study distrrict, 36 (31 %) were each from Bolori and Gwange, 39 (33.6 %) from Kyarimi Park and 5 (4.3 %) from Shehuri North. 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 Park and Shehuri North respectively (Figure 2). There was no significant difference (P=0.095) among the clusters. Out of the 284 egg samples collected from the retail outlet, 201 (70.1 %) samples were from Jos and 83 (29.2 %) from Ibadan. A total of 100 (35.2 %) samples were positives for antimicrobial screening which comprises of 71 (35.32 %) and 29 (34.94 %) from Jos and Ibadan respectively. with no significant difference between the two sources (P=0.902). From this study it was concluded that: There is small flock size (back yard) farm in Maiduguri with 94.3 % of the farmers holding equal or less than 500 birds in their farms. High percentage of antibiotic contamination was detected in the study area.

Key words: table egg, *Bacillus cereus,* Maiduguri Metropolis, Antimicrobial residue

Introduction

Egg contain carbohydrate, protein and other essential substances required for human existence (Braun, 2000). The low caloric value, edibility and nutrient content makes egg significant food stuff for many dietary regimes (Kenner *et al.,* 2006). The hen's egg is a 'self-contained unit for starting a new life (FAO, 2013). The egg is a major food source, providing good quality balanced nutrient to billions of people throughout the world, the world's total 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 important component of Nigerian 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 occupies a prominent position in the provision of animal protein and this account for about 25% of local meat production in Nigeria (Agbaje *et al.,* 2010). The annual production capacity of commercial eggs in Nigeria is estimated at 8, 216, 208, 000 eggs equivalent to 273, 873, 600 crates of eggs (FAO, 2008). 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 disease outbreaks (Al-Ghamdi *et al.,* 2000). They are used by the poultry industry to enhance growth, feed efficiency and reduce bacterial disease (Donoghue, 2003). In layer hens, antimicrobials are only used to treat and prevent bacterial infections. Some of the antimicrobial classes used in treating layers include aminoglycosides, tetracyclines, beta-lactams, quinolones, macrolides, polypeptides, amphenicols and sulphonamides (Stolker and Brinkman, 2005). Through the years the issue of antibiotic residue from farm animals and their effect on human health has been a major concern (Bahry *et al.,* 2013).

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

2006; Castanon, 2007). In Maiduguri, the study area, antibiotics are freely marketed without veterinary prescription (Geidam *et al.,* 2012) and despite 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.

MATERIAL AND METHOD

Study area

Maiduguri Metropolis, a major city in the Northeastern part of Nigeria, is located between 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^2, which makes it the largest city in the Northeastern region of Nigeria (Daura, 2002; Jimme *et al.,* 2016). Maiduguri city now extends to four Local Government Areas: Maiduguri Metropolitan, Jere, Konduga and to a smaller extent part of Mafa local 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 remaining part of the year (Jimme *et al.,* 2016). There are four identified seasons in the area which include the *Rainy Season*, (June to September) *Harvest Season* (September to 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 2006 census (NPC, 2008). With an annual growth rate of about 3.5% and a density of 1145 persons per square km which makes it the most densely populated city in North Eastern Nigeria (Waziri, 2009; Jimme *et al.,* 2016). Crop production and livestock farming are the 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).

Study design

Sampling technique

Multi stage sampling method was used for sample collection.

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

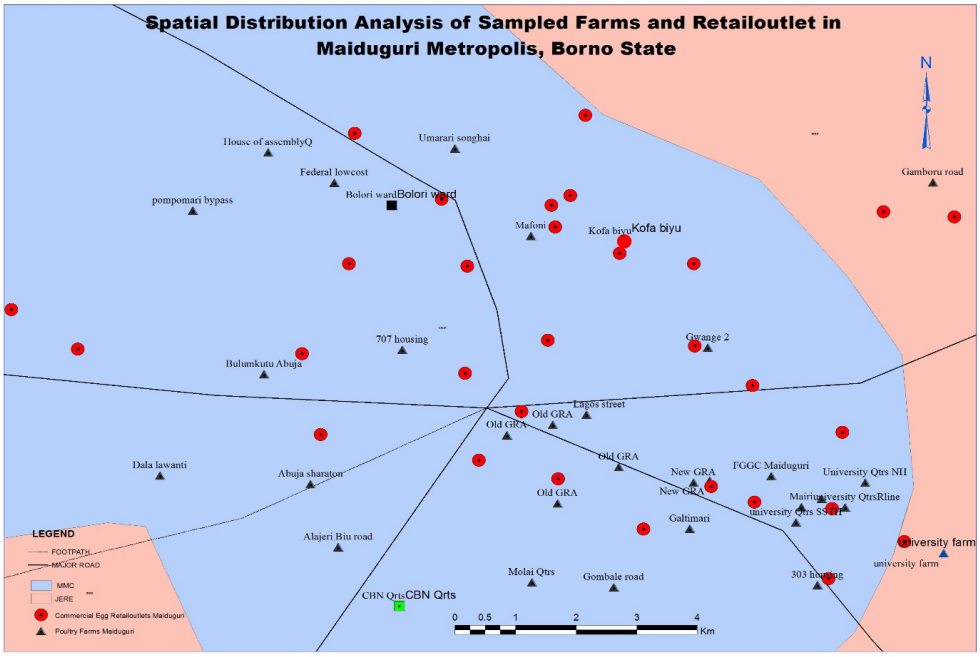


FIGURE : 1

Sample size determination

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

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

Where:

n= sample size

z=desired confidence 1.96

p=prevalence= 3.6 % by fagbamila et al., (2010) in Jos plateau state.

q= 1-p

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).

Sample collection

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

Sample processing

The antimicrobial screening of eggs was carried out using the disc diffusion method where *Bacillus cereus* ATCC 14579 from spectra medics' laboratory in Ogun state was used as the 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 surface was thoroughly cleansed using sterile cotton wool soaked in 70% alcohol. Sterile forceps were used to puncture the egg at the tip to create a small opening from where the yolk were carefully drained out into a sterile beaker and mixed with Phosphate Buffer Saline pH 7.4 and then thoroughly homogenize, then 10 milliliter was transfer to a clean sterile test tube and was centrifuge for 10 minutes at 4000 x g. Two milliliter of the supernatant was transferred to sterile petri dish (Fagbamila *et al.,* 2010; Kabir *et al.,* 2004).

Qualitative screening

Using a clean sterile forceps Whatman® filter paper disc 0.6 cm in diameter was dipped into 2 mm of the egg supernatant in the Petri dish, until it is soaked and then were exposed to temperature of 80^0C for 10 minutes to in activate inhibitory substance and placed gently on 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 37^0C for 24 hours after which the plates were viewed for the presence or absence of zones of inhibition of the test organisms around discs. Any disc with a zone of inhibition greater than 1 mm around the disc was considered positive (Kabir *et al.*, 2004).

126 **Data analyses**

127 The data was compiled and analyzed 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 **Result**

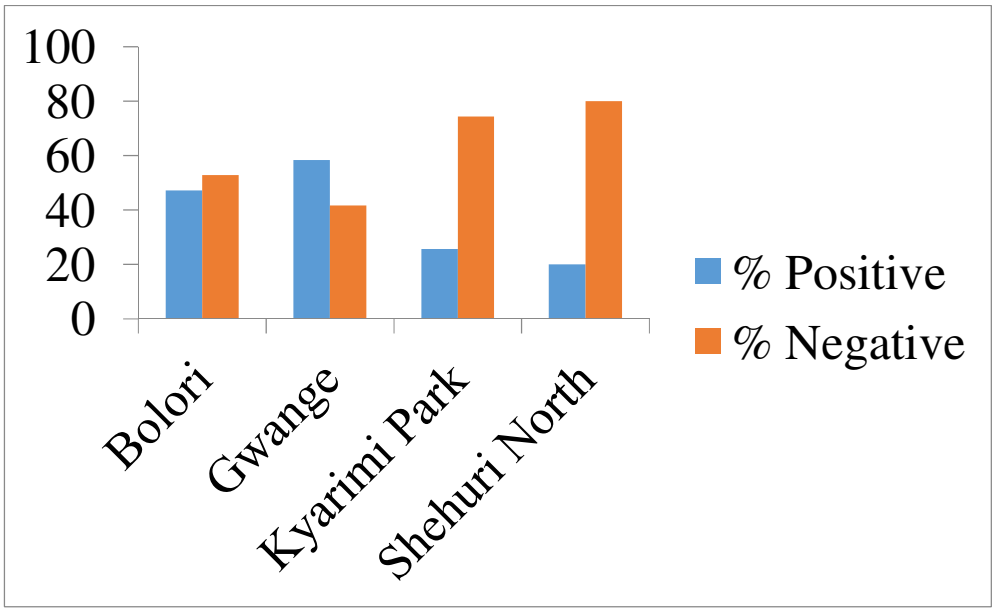
131 One hundred and sixteen (116) table eggs collected from farms across the study area, 36 (31
132 %) were each samples from Bolori and Gwange farms, 39 (33.6 %) from Kyarimi Park farms
133 and 5 (4.3 %) from Shehuri North farms. A total of 49 positive samples were obtained which
134 include 17 (47.2 %), 21 (58.3 %), 10 (25.6 %) and 1 (20 %) from Bolori, Gwange, Kyarimi
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
139 (34.94 %) from Jos and Ibadan, respectively (Figure 3) with no significant difference of
140 residue of antimicrobials between the two sources (P=0.902).

141

142

143

144

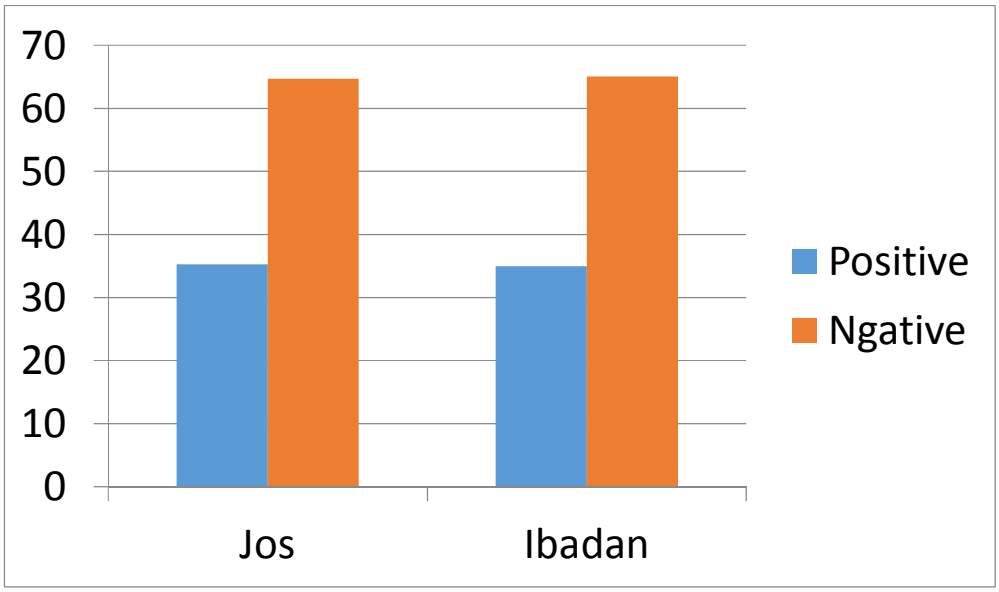


145

146

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

148 **Poultry Layer farms in Maiduguri Metropolis, Nigeria**



149

150 **Figure 3: Result of qualitative screening for antibiotics residues from retail outlet in**

151 **Maiduguri Metropolis, Nigeria**

152

Discussion

All the samples used for the study were obtained from layer farms and retail outlets. The majority of the retailers source their eggs from Jos and Ibadan in order to compensate for the short fall from local production in the 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 in Maiduguri Metropolis. Antibiotic residues were detected in 49 (42.2 %) of the eggs samples collected from farms with lower percentage 100 (35.2 %) in egg samples collected from retail outlets. The lower percentages in retail outlets may be due to storage or variation of antibiotic use by different farms were the eggs were sourced. This is in tandem with observation of Ezenduka *et al.,* (2011) in Enugu who reported 36 % positive in eggs sampled from farms and 30 % in retail outlets and El-Nasri *et al.,* (2012) also reported 55.4 % antibiotic residue in eggs collected from farms and 43.2 % in retail outlets. Kabir *et al.,* (2004), Fagbamila *et al.,* (2010) and Omeiza and Nafarnda, (2015) reported 0.5 %, 3.6 % and 18.5 % antibiotic residue in table eggs respectively. This might not be unconnected with variation in awareness of biosecurity, antibiotic residue in table eggs and public health effect of antibiotic residue.

Conclusion

From this study it was concluded that: There is small flock size (back yard) farm in Maiduguri with 94.3 % of the farmers holding equal or less than 500 birds in their farms. High percentage of antibiotic residue was detected in the study area, with 42.2 % and 35.2 % positive in table eggs collected from layer farms and retail outlets respectively.

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

177 sensitive techniques should be carried out to quantify the residue levels of individual
178 prohibited for used in food producing animals. Antibiotics Legislation regarding the use of
179 prohibited antibiotics on food animals by National Agency for Food and Drug Administration
180 and Control.

181 **Reference**

182 Agbaje M, Davies R, Oyekunle MA, Ojo OE, Fasina FO and Akinduti PA (2010).
183 Observation on the occurrence and transmission pattern of *Salmonella gallinarum* in
184 commercial poultry farms in Ogun State, South Western Nigeria**.** Afr. J. Microbiol
185 Res. 4(9): 796-800.

186 Al-Gamdi M, Almustafa Z, El-Morsy F, Al-Faky A, Haider I, Essa H (2000). Residues of
187 tetracycline compounds in poultry products in the eastern province of Saudi Arabia.
188 Public Health. 114: 300-304.

189 Bahry SN, Mahmoud IY, Al-Musharafi SK (2013). The overuse of tetracycline compounds in
190 chickens And Its Impact on human health. J. Ala. Acad. Sci. **77:** 152-159.

191 Braun P (2000). Freshness of table eggs during storage. Worlds Poult. Sci. J. 16(10): 41-41.

192 Castanon, J. I. R. (2007). History of the use of antibiotic as growth promoters in European
193 poultry feeds. Worlds Poult. Sci. J. 86: 2466–2471.

194 Daura MM (2002) “Maiduguri” Atlas of Nigeria in Africa Atlasses. Bietlot, Belgium. 148-
195 149.

196 Daura MM, Gisilanbe AM, Waziri M (2001). Flood Plain Encroachment and Hazard
197 Awareness in Urbanized Catchments: A Study of Ngadda Flood Plain in Maiduguri.
198 In: Daura.

199 Donoghue DJ (2003). Antibiotic residues in poultry tissues and eggs: human health concerns.
200 Worlds Poult. Sci. J. 82: 618–621.

201 El-Nasri A, Salman M, Osman, AM (2012). Detection of antibiotic residue in table eggs
202 using disc assay and premi test in Khartoum State, Sudan. J. Vet. Med. Anim. Pro.
203 3(2): 16-27.

204 Ezenduka EV, Oboegbulem SI, Nwanta JA Onunkwo JI (2011). Prevalence of antimicrobial
205 residues in raw table eggs from farms and retail outlets in Enugu State, Nigeria.
206 Trop. Anim. Health. Prod. 43(3): 557-559.

207 FAO (2008). Assessment of the Nigerian poultry market chain to improve biosecurity. Pp:10-
208 11.

209 FAO (2013). Improving the safety and quality of eggs and egg products, Vol 1; Egg
210 chemistry, product and consumption. Wood head publishing series in food science,
211 technology and nutrition.

212 Fagbamila I, Kabir J, Abdu P, Omeiza G, Ankeli P, Ngulukun S, Muhammad M, Umoh J
213 (2010). Antimicrobial screening of commercial eggs and determination of
214 tetracycline residue using two microbiological methods. Int. J. of Poult.Sci. 10: 959.

215 Geidam YA, Ibrahim UI, Grema HA, Sanda KA, Suleiman A, Manzo DL (2012). Pattern of
216 antibiotic sales by Drug stores and usage in poultry farms: a questionnaire-based
217 survey in Maiduguri, North eastern Nigeria. J. Anim. Vet. Adv. 11(16): 2852-2855.

218 Hofacre CL (2006). Antimicrobial drug use in poultry.In: Antimicrobial Therapy in
219 Veterinary Medicine. (Eds Giguere S, Prescott JF, Baggot JD, Walker RD, Dowling
220 PM,) pp. 545–554. Blackwell Publishing, Ames, IA.

221 Jimme MA, Bashir A, Adebayo AA (2016). Spatial distribution pattern and Terrain analysis
222 of urban flash floods and inundated areas in Maiduguri metropolis, Borno state,
223 Northeast, Nigeria. JGIS. 8: 108-120.

224 Kenner K M, McAvoy KC, Foegeding JB, Curtis PA, Anderson KE, Osborne JA (2006).
225 Effect of testing temperature on internal egg quality measurement. Worlds Poult.
226 Sci. J. 85: 550-555.

227 NPC (2008) National Population Commission. 2008. Maiduguri Projected Population.

228 Omeiza GK, Nafarnda WD (2015). Annual trend in the occurrence of antimicrobial drug
229 residues particularly chloramphenicol using a comparative detection methods in
230 Federal Capital Territory (FCT), Abuja, Nigeria. J. Environ. Sci. Toxicol. Food
231 Technol. 9(9)*:* 60-66.

232 Sirdar MM, Picard J, Bisschop S, Alexander R, Jambalang BG (2012). A survey of
233 antimicrobial residue in table eggs in Khartoum state, Sudan. J. Vet. Res. 79(1):1-9.

234 Stolker AA, Brinkman UA (2005). Analytical strategies for residue analysis of veterinary
235 drugs and growth-promoting agents in food-producing animals--a review. J.
236 Chromatogr. A. 1067:15-53.

237 Thrusfield M (1995). Veterinary Epidemiology 2nd ed. Blackwell Science Ltd, Oxford. pp.
238 296–311.

239 Thrusfield M. (2005). Diagnostic testing. In: *Veterinary Epidemiology*, third ed. Blackwell
240 Science, Oxford, DQ, UK, pp. 305–330.

241 Tijjani H, Tijani BA, Tijjani AN, Sadiq MA (2012). Economic analysis of poultry egg
242 production in Maiduguri and environs of Borno State, Nigeria. SJAS. 2(12): 319-
243 324.

244 Waziri M (2009) Spatial Pattern of Maiduguri City: Researchers' Guide. Adamu Joji
245 Publishers, Kano City.