Low Egg Production Performances Associated With Multi-drug Resistant Bacteria Among Some Nigerian Commercial Layer Flocks

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Abstract

Bacteriological investigations were carried out on commercial layer flocks of 19 poultry farms in parts of Lagos and Ogun States of Nigeria, in the years 2005 and 2006. The layers totalling 21,459 (range 1,050 – 2,900) had records of low egg production performances. Cloacal swabs and blood from live birds, as well as liver, spleen and heart blood of dead birds were investigated for pathogenic bacteria. The isolated pathogenic bacteria include Escherichia coli, Salmonella enterica serotype Typhimurium, Salmonella enterica serotype Enteritidis, Salmonella enterica serotype Gallinarum, Klebsiella aerogenes and Klebsiella aerogenes. The bacteria were resistant to multiple of drugs. Administration of antibiotics following laboratory investigations, as well as regulations to the use of antibiotics and other medicaments are highly necessary for the growth of the Nigerian poultry industry.

Key words: Bacteria, drug resistance, layers, low egg production, poultry.

Introduction

Intensive animal production is associated with large scale antimicrobial use worldwide (Moulin, 2001). This often leads to a high level of colonization of animals with antimicrobial resistant bacteria (Corpet, 1988; Aubry-Diazon et al., 2004). Infections due to such drug resistant organisms are of grave consequences as they can lead to high economic loss in poultry (Puppe et al., 1991; Ojo, 1991). Ojeniyi (1989), has long time reported higher incidence of drug resistant bacterial isolates obtained from commercially reared poultry than isolates from locally reared birds in Ibadan, Western Nigeria.

In Nigerian poultry industry, the use of antibiotics is without restriction. Often, farmers themselves and quacks administer drugs on flocks without supervision by veterinarians. Low egg
Drug resistant bacteria and low egg production

production performances among commercial
layers could be due to factors such as injections,
nutrition, stress and management lapses among
others (Palmer, 1991; Sprightly, 1993). However,
when there is a drop in egg production most
Nigerian farmers go for antibiotics. If he uses
one antibiotic preparation without result, he
changes to another until he is able to get result.
The article presents a field report of
investigations carried out on some commercial
layer flocks with complaint of low egg
production performances and measures taken
to resolve the problems.

Materials and Methods

Ten commercial poultry farms located in some
towns and villages in parts of Lagos and Ogun
States, Nigeria, were investigated for the cause
of drop in egg production among their layers
flocks in the years 2005 and 2006. The
commercial layer flocks of the ten poultry farms
investigated for bacteriological cause of drops
in egg production were 45.0 - 18.3 weeks old
(mean ± SD) and 21,450 (range 1,050 - 2,900)
in total.

The birds in the farms investigated had
records of low egg production performances with
occasional low mortalities. The farms’ records
were gone through, birds were closely examined,
the buildings inspected and post-mortem
examination carried out on dead birds. On the
spot investigations eliminated some problems
commonly associated with drop in egg
production such as: accommodation, feeding,
watering, Newcastle disease (absence of typical
clinical signs and pathognomonic lesions), as
well as worm (based on examination of facial
droppings and intestinal contents) and
coccidial infections (absence of fleas and
lice).

Bacteriological Investigations

For each of the ten farms, cloacal swabs were
drawn from 20 birds and blood samples (0.5-
1.0ml) were collected via the wing veins with
the aid of syringes and needles from another 20
birds. All the birds bled were randomly selected.
Livers, spleens and hearts were harvested from
dead birds obtained from the farms and taken to
the laboratory in sterile bottles. A loopful of
cloacal swab rinsed in sterile distilled water,
blood samples, fluids from liver and spleen as
well as blood from heart were streaked
separately on freshly prepared MacConkey agar
plates. The agar plates were incubated at 37°C
for 18-24 hours. The bacterial isolates were sub-
cultured in selective media, subjected to
biochemical tests, characterized and identified
according to Barrow and Feltham (1995).

Antibiotic susceptibility tests (AST) on the
isolated bacteria were determined by the disk
diffusion technique (NCCLS, 1997). The
antibiotics and their concentrations in
micrograms (µg) were as follows: Augmentin
30, Ofloxacin 15, Tetracycline 25, Gentamycin
10, Amoxicillin 25, Cefuroxime 20 and
Nitrofurantoin 20. K. pneumoniae (NCTC 10418)
served as control.

Based on the degree of clearance from the
antibiotic disk, the results obtained were
classified as highly susceptible (> 10mm),
intermediately susceptible (< 10mm) or resistant
(no clearance), which is the standard being used
by the University of Benin Teaching Hospital.
Treatment of Birds

Brokers were administered the drugs based on the outcome of the AST performed. Birds in forms A, E, F, and G were treated with oral preparation of Conflora™ (Concept Pharmaceutical Ltd., Mumbai, India), an enrofloxacin, for five days. Birds in forms B, C, D, H, and I were treated with oral administration of pure Furaltadone™, a nitrofurantoin, for a period of five days at a dosage of 0.5g/litre. Eggs production performances before medication and 3 weeks post medication with appropriate drugs were recorded. Average of one week production was considered for each flock.

Results

The bacteria isolated from the commercial laying flocks of the ten farms investigated for bacteriological cause of low egg production performances included, *Escherichia coli* (33%), *Salmonella enterica* serotype Typhimurium (8.33%), *Salmonella enterica* serotype Enteritidis (16.67%), *Salmonella enterica* serotype Gallinarum (8.33%), *Klebsiella aerogenes* (8.33%), and *Klebsiella oxytoca* (16.67%) (Table 1). The distribution of the bacterial isolates in the investigated organs, harvested from

and resistant = 0. Student's t-test was used to determine the statistical difference in percentage egg production performances of the flocks before and after medication.

<table>
<thead>
<tr>
<th>Form</th>
<th>Isolated bacteria</th>
<th>Aus</th>
<th>Ofl</th>
<th>Tol</th>
<th>Gen</th>
<th>Ame</th>
<th>Cot</th>
<th>Nit</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>B</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>C</td>
<td><em>Salmonella Enteritidis</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>D</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>E</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>F</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>G</td>
<td><em>Klebsiella aerogenes</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>H</td>
<td><em>Escherichia coli</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>I</td>
<td><em>Salmonella Enteritidis</em></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>J</td>
<td><em>Salmonella Gallinarum</em></td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
</tbody>
</table>

Average score: 2.38
Percentage susceptibility: 70.1%
Percentage resistance: 0%

Key:
A - Amoxicillin
O - Oxytetracycline
T - Tetracycline
C - Cefotaxime
N - Nitrofurantoin

Table 1: Antibiotic susceptibility pattern of isolated bacteria

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Table 2: Distribution of bacterial isolates in body organs.

<table>
<thead>
<tr>
<th>Farms</th>
<th>Bacteria</th>
<th>Venous blood n=20</th>
<th>Liver n=30</th>
<th>Spleen n=10</th>
<th>Heart blood n=10</th>
<th>Clinical swabs n=20</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td><em>Escherichia coli</em></td>
<td>75</td>
<td>130</td>
<td>90</td>
<td>100</td>
<td>55</td>
</tr>
<tr>
<td>B</td>
<td><em>Escherichia coli</em></td>
<td>ND</td>
<td>100</td>
<td>100</td>
<td>90</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td><em>Salmonella Enteritidis</em></td>
<td>80</td>
<td>90</td>
<td>90</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>C</td>
<td><em>Escherichia coli</em></td>
<td>ND</td>
<td>80</td>
<td>70</td>
<td>80</td>
<td>ND</td>
</tr>
<tr>
<td>D</td>
<td><em>Klebsiella aerogenes</em></td>
<td>ND</td>
<td>70</td>
<td>70</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>E</td>
<td><em>Salmonella Typhimurium</em></td>
<td>85</td>
<td>90</td>
<td>80</td>
<td>70</td>
<td>50</td>
</tr>
<tr>
<td>F</td>
<td><em>Escherichia coli</em></td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>ND</td>
</tr>
<tr>
<td>G</td>
<td><em>Klebsiella aerogenes</em></td>
<td>ND</td>
<td>60</td>
<td>70</td>
<td>70</td>
<td>ND</td>
</tr>
<tr>
<td>H</td>
<td><em>Escherichia coli</em></td>
<td>75</td>
<td>90</td>
<td>90</td>
<td>80</td>
<td>100</td>
</tr>
<tr>
<td>I</td>
<td><em>Salmonella Enteritidis</em></td>
<td>ND</td>
<td>90</td>
<td>80</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>J</td>
<td><em>Salmonella Gallinarum</em></td>
<td>50</td>
<td>70</td>
<td>50</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

Values within table are in percentages.

The bacteria indicated from eteology were isolated along with many intestinal flora.

diseased birds, were as presented in Table II. The prevalence of pathogenic bacteria species among the laying flocks of the ten farms is illustrated in Figure 1: *Escherichia coli* being the most prevalent. The bacteria were resistant to multiple of drugs. All the bacterial isolates were highly susceptible to nitrofurantoin and augmentin, but with varied degrees of resistance to the remaining five antibiotics used (Table I).

Egg production performances before medication and 3 weeks post medication were as presented in Figure 2. Treatment with appropriate drugs improved egg production significantly (F = 2.41, P < 0.05).

**Discussion**

One of the most common problems in breeders and commercial layers is investigating the cause of drop in egg production (Pattison, 1993). Drop in egg production is associated with anxiety on the part of the farmer, as he is being desperate to the return production to peak often abuse antibiotics usage.

In Nigeria, farmers having commercial layer flocks with stocking capacity over 3,000 birds are many. They may contribute up to 50% of total eggs produced in the country. Majority of such farmers do not engage the services of veterinary surgeons, as they consider such services too expensive. Some engage the services of veterinarians as consultants when commencing the poultry project, but after one or two years they believe they have acquired enough knowledge and terminate such services. In summary, most Nigerian farmers in this category administer drugs to flocks by themselves or patronize quacks whose services they considered less expensive. They seek for assistance from
qualified veterinarians only when they might have tried their best without achieving the desired results (Okiki, unpublished field observations). All the farmers reported in this study had stocking capacities less than 3,000 commercial layers. The farmers admitted that they had used at least 2 different types of antibiotic preparations before our intervention. The resistance to many drugs by the isolated bacteria from these flocks (Table 1) attested to the fact that antibiotics had been abused in the farms. The significant increase in egg production (Figure 2), following treatment of flocks with sensitive drugs (drugs selected from AST results, Table 1), is an indication that inappropriate drugs were earlier administered. Earlier studies in these locations reported the prevalence of multi drug resistant bacteria in poultry farms (Okiki et al., 2006) and association of multidrug resistant gram-negative bacteria with high mortalities among commercial poultry flocks (Okiki and Ogbum, 2008).

The isolated bacteria in this study might have acquired resistance to multiple drugs as a result of usage of such drugs at a dosage below the curative level on the farms. The drugs used could have suppressed the bacterial infection from developing into disease state. This stage of infection with bacteriaemia could produce stress in the birds which could be responsible for such drops in egg production.

Multidrug resistant gram-negative bacteria has been emerging worldwide (Yamane et al., 2005) and resistance to antibiotics could be plasmid or chromosomal mediated (Mayer, 1988). Diagnostic uncertainty has however been reported as a key driver of drug misuse and overuse, which can lead to antimicrobial
selection pressure and increased rates of resistant microbes (Kuernberger and Bishai, 2004). Fischer et al. (2004) noted that the risks associated with untreated microbial infections and the lack of accurate clinical and laboratory-prediction methods result in low threshold for initiating empirical antimicrobial drug therapy. In Nigerian poultry industry, laboratory investigations of etiology of diseases are not yet popular as diagnosis of diseases are still based on physical observation of flocks and autopsy which are insufficient. The future of Nigerian poultry industry should depend on improved diagnostic techniques for microbial infections.

Whenever there is drop in egg production, use of antibiotics should not be considered first, rather the cause of low performances should be determined. In investigating the cause of drop in egg production among breeders and commercial layer flocks, it is recommended that the under listed points should be checked as described by Pattison (1997):

(a) Water consumption: has there been a sudden interruption in water supply? Check water meters, if fitted in each house. Water samples should be taken for investigation of possible contamination (especially if water supply is uninterrupted).

(b) Does the drop in production coincide with a new feed delivery and if so has this batch of feed been distributed to the birds in all the houses? A feed sample should be taken from current feed batch and should be analysed for any major abnormalities, initially to ensure it is the correct feed i.e. layer or breeder feed.

Then one can assay for protein, energy, calcium and mycotoxin levels.

c) Are the poultry well ventilated? If not necessary correction should be done.

d) Do the birds look healthy? If not, are there signs of diseases or a rise in mortalities?

e) Are the eggs normal in shape and size? If not, are there soft shells or excess calcification?

(f) Has there been a change in management or staff looking after the birds? Also, has there been visitors who could have frightened the birds e.g. a vaccination team?

g) Is the stocking density on the floor or cages appropriate to the breed and climate?

(h) Check point of lay weights and rearing history and vaccination records.

(i) In case of mortalities, take a selection of organs of organs to store in deep freezer for later bacterial isolation if necessary e.g. trachea, liver, spleen, caecal tonsil and gut contents.

(j) Take blood samples from affected houses to be tested for infectious diseases such as Newcastle disease, avian influenza, infectious bronchitis, etc.

**Recommendations**

In order to reduce the incidence of multi-drug resistant bacteria and their socio-economic effects in the poultry industry, the following are hereby recommended:

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1. Strict bio-security will reduce bacteria and other infectious microorganisms in the poultry confinement.

2. There should be regulations to the use of antibiotics in poultry so as to prevent further abuse of antibacterial agents.

References


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