Micro-organism Pollutants Associated with Poultry Feed from Three Different Companies

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Abstract
The objective of this study is to ascertain the microbial safety of commercially prepared Poultry feeds produced by companies. The type of starters mash from three different companies (i.e. Top feed, vital and Guinea feed) was used in the research. The samples were collected with sterile spatulas from 150 different bags in the ratio of 50 500 and were promptly transferred to the laboratory for analysis. The microorganism counts obtained in this study were high and indicated the need for improve sanitary conditions in the factories and farms. The results indicated that hazard Analytical critical control point (Haccp) should be observed during the harvesting of the raw materials, processing, packaging and storage of the finished product. Good trained personnels should be employed to work in Poultry feed factories to ensure well processed Poultry feed.

Introduction

Poultry are collection of birds raised commercially or domestically for meat, egg and feathers. Chickens, ducks' turkeys and geese are of primary importance while guinea fowl and squabs (pigeons) are chiefly of local interest. These birds are source of income and food to the rarer. In the past only hens that could no longer produce eggs were killed and sold for meat but by the mid-20th century, meat production had outstripped egg. Production as a specialized industry (EncycloparrediaBritaimca, 1988). Heavy breed Poultry animals are used for meat white light bread chickens are primarily for the production of eggs (Mc Graw Hill encydopaedia 1992). The meat and eggs produced from Poultry are important sources of dietary portion. In Africa, animal protein foods from all sources contribute between 7g and 15g daily per person to the total food intake (Oyenyga, 1974). Today, Poultry production in the country is increasing at a tremendous rate and yet cannot meet the demand because of population growth and the awareness created by the campaigns for the need of animal protein in the diet.

The growth and well-being of organisms are to a large extent dependent on the mount and type of food they receive and the manner or rate at which they receive it. The food should contain nutrients such as water, carbohydrates, fats, protein, minerals, vitamins and added growth factors improper balance. Each of these nutrients has a major role to play concerning growth, maintenance and productivity of the Poultry. According to Leonard 91981), nutritional deficiency or inbalance. These diseases may include rickets, perosis, and nutritional roup, curly the paralysis etc. These diseases could result due to deficiency in vitamins D, Manganese, vitamin A, vitamin E and riboflavin. Improper sanitation and inadequate management play major roles in disease. Outbreak and productivity of the desired products. In developing countries like Nigeria, the cost of feeding, especially the monogastries continues to escalate primarily in response the soaring cost of the conventional feed ingredients. This phenomenon is precipitated by under production and importation of some food stuff coupled with the competition between man and farm animal for energy and protein concentrates. The use of agro-industrial by-products towards reducing food cost has been widely recognized (EshieH and Ademosun, 1981; Atteh and Oloagbenla, 1993). Chicken and turkeys satisfy their energy needs provided the ration a allows them to do so (Church, 1988). Theare of course exceptional to this rule particularly where heavy breed layers are concerned, when birds have a tendency to over-eat. Where this is a problem, it is general practice to subject the birds to some degree of food restriction during the growing and production periods. The major ingredients that are integral parts of Poultry rations at the present time in the USA are corn, as the primary source of energy and soybean meal as the major protein supplement (Roland et al 1972), while in Nigeria, Udedibie et al (1988) and Nwokon (1993) produced Poultry offal meal (P.O.M) and chicken offal meal (COM) respectively from waste product similar to Poultry visceral Offal (PVO) according to these authors POM has been used to replace ground nut cake in layer and broiler finisher diets and COM has been used along with Poultry meat and blood meat as sources of methodize and lysine in the starter diets of lockerels. These major ingredients, usually available in plentiful supply, allow rapid growth or high egg production with very efficient conversion. However, corn-soy rations are deficient in some nutrients for chickens and these nutrients are normally supplied by other Poultry meal. There are different types of feeds given to Poultry brids depending on the purpose. These different types of feeds given to Poultry birds depending on the purpose. These different types are the breeders' starters, growers' and layer' feeds. The percentage of each ingredient in the feed varies depending on the type of feed. The ingredient includes ground yellow corn, ground oats or barley, Alfata meat, Poultry meal, Oystershell, managised salt, vitamin A, vitamin D, and coccidiostat (which is given in form and at the level recommended by the manufacturer). The different percentage of each ingredient varies depending on the kind of feed for example, ground yellow corn in starter and grower feed is 25% and 22% in breeder and layer feeds. There could be the same percentage of a particular ingredient in different feeds as in the case of a alfafa meal which constitutes 10% inall the feed types. Some ingredients could be absent in a particular feed but present in the others as in the case of riboflavin supplement which is absent in starter feed and present at the levels of 2 and 5% in rower, layer and breeder feeds respectively.

To maintain healthy birds, the feeds are kept fresh as much as possible at all times. The amount of feed in feeders are limited to the extent necessary to avoid wastage. It is a good practice to fill hanging feeders – only three quarter full, and trough feeders only two third full (Graham, 1977). Checking the weight of the birds and its feed consumption is very necessary. A drop in feed intake usually is the first indication of trouble, a disease outbreak, molt, stress or poor management (Graham, 1977). Most Poultry feed are prepared in dehydrated forms and because of this, there is the need to store them properly to avoid moisture uptake and damage by heat. In this way, They can remain safe for a considerable period of time without losing their safeness and value. The feed should be stored in a suitable place where it will not be attacked by microorganisms, insects, rodents, etc. Air tight storage is not advisable because offensive odour might result when there is obstruction of out flow and inflow of air (Leonard, 1981). High temperature and oxidation destroy certain vitamins; therefore, care must be taken in the preservation and storage of feeds to protect the vitamins they contain (Mc Graw – Hill encyclopaedia 1992). The condition of feed storage and handling could be a source of contamination. When feeds are unhysgenically handled and stored, there could be a buildup of microbial pollutants. Poultry have been found to be susceptible to infection due to certain microorganisms and man may be secondarily infected through heavy contaminated food such as Poultry meat and eggs (Gorden and Tucker, 1965, Hall, 1977, and Barrell, 1982). Micro-organism that may contaminate feed include the following general another disease caused by salmonella infections is paratyphoid. Paratyphoid is an infection disease of chi,Entherobacter,Escherichia, Protens,Pseudomonas,staphylococcus, salmonella, shigella, providencia,serratia,Klebsiella, streptococcus, clostridium, Aspergillus and Erysiphelothrie(Leonard, 1981). Typically, in some countries, about 50% of all outbreaks of salmonellosis in man are caused by infections derived from these sources and the incidence of salmonella in Poultry carcasses has, in certain cases been found to be 7% or more (Van schothorstNotermans, 1980). Okongi (1984) examined Poultry feed and found that salmonella was present in the sample. Another disease caused by salmonella infections is paratyphoid, Paratyphoid is an infections disease of chicken, turkeys, ducks and other birds. Bofulism another kind of disease occurs in both young and adult Poultry birds. It is caused by a bacterium, Clostridium Botulinum. This organism grows in decaying plants and animals' materials. Birds feeding material containing the toxins produced by the microorganism lose control of their neck muscle (Leonard, 1981).

Methods

Sterilization

Already, properly washed test tubes, petri-dishes, pipettes, conical flasks, beakers, bottles, wire loop, spatulas and inoculating needles, were sterilized in hot air oven at 160^0c for one hour in their respective canisters and stored at 41^0c.

Source of Samples

The type of Poultry feeds used in the research were starters mash from three different companies (i.e Top feed, vital and Guinea feed). The starters mash was collected with sterile spatulas from

150 different bags in the ratio of 50¨500 and were promptly transferred to the laboratory for analysis

Preparation of Culture Media

All the media used for culturing were aseptically prepared accoring to the manufacturer's instructions and autoclaved at 121^0c for 15 mins at 15165/Ps1. The media include nutrient agar and MacConkey
agar.

Determination Of pH

The pH was determined using the method described by Ofunya and Akpoti (1988). Lg sample of Poultry feed was suspended into 30ml of demonized neater. It was stirred and allowed to settle for 1 minutes before the pH was determined with a pH meter (Table 3).

Plating Technique

The "streak plate" method was employed. A loopful of suspension was removed aseptically, from each of the samples, using a sterile wire loop. Then the loop was streaked very gently over the surface of the solidified Nutrient agar. Care was taken so that there was no cut into the agar with the wire loop. The streaking was done in a characteristic patterns, in which the loop was drawn over a surface of the agar with three streaks. The loop was flamed another three parallel streaks made over area B and latter C area, and completed with a snake – like streak towards the centre of the plate. The plates were covered, inverted and incubate for 24 hours at 37^0c

Bacteri Count, Gram Staining and Microscopic: -

i. microorganism Count: -

After incubation the number of colonies on the petri-dishes were counted using the colony counting chamber. The average total and differential standard plate counts (SPC) were taken (Table 1 and 2 respectively).

ii. **Gram Staining and Microscopic Work:**

Grown colonies were gram-stained using representatives of the different groups. Later the stained smears were examined microscopically. Later representative colonies were separately sub-cultured for confirmatory tests.

Biochemical Test for Identification of Microorganism Isolates.

A Catalase Test

This test was performed according to the method of Barker and silverton (1985). With the aid of a glass rod, a portion of the microorganism colony was transferred on to a clean slide. Two drops of hydrogen peroxide were placed on the organism and left for a few minutes. Production of gas bubbles indicated a positive reaction.

B. Indole Test

The peptone water medium was inoculated and incubated for 48 hours at 37^0c. the tube was further allowed to stay for more 48 hours in the incubator for the accumulation of indole. After this period, 0.5ml kova's reagent was added separately to each tube and swirled gently. The appearance of a red colour in the alcohol layer indicated a positive reaction.

C. Methyl Red Test

With the aid of a sterile were loop, the MRVP medium was lightly inoculated with the test isolates and incubated at room temperature for 2 days. After the incubation period, 5 drops of methyl red reagent were dropped into each culture tube. Production of red colour immediately indicates a positive reaction while negative were yellows.

D. Voges-Proskaver Test

Sterile medium used was inoculated and incubated at 37^0c for 48 hours. One ml of 40% KOH and 3ml of 5% alpha-nepthol in absolute alcohol (ethanol) were added. A positive test was shown by the development of pink colour in 2-5 minutes, which would become crimson in 30 minutes. Colour in 2-5 minutes which would become crimson in 30 minutes.

E. Oxidase Test

The wet filter paper test, Baker and Silverton, (1985) was utilized for this test. The test depends on the presence in microorganism of certain oxidases that will catalyse the transport of election donors' ins the microorganism and a redoz dye tetraethyl -p-Phenyhenediamines. Two or three drops of the oxidise reagent were placed on a piece of filter paper. With a glass rod and left for 5-10 seconds. In a positive result, the dye is reduced to a deep purple colour and no colour change is a negative result.

F. Citrate Utilization Test

A small amount of the culture growth was picked from the Nutrient agar plates, using glass rod and highly inoculated on simon citrate agar and incubated at 37^0c for 24 hours. A positive test was indicated by the development of a blue colour in the medium. While in a negative reaction the medium retained its original pale green colour after incubation.

G. Hydrogen Sulphide Production/Sugar Fermentation

A sterile needle was used to inoculate each organism into TS1 agar slant by stabbing the agar tube to the bottom with the inoculum and streaked across the top of the slant. The tubes were incubated at 37^0c for 48 hours. When only glucose is fermented the slant will be pink and negative will be yellow, when sucrose and or lactose is fermented, the slant will turn yellow. Gas cracks in the agar hydrogen sulphide production is seen by the blackeing of the medium.

H. Motility Test

About 2-3 drops of peptone water with growth of the organism was placed on a clean slide with a loop. The cover slip was placed over the slide the slide was left for some time and them examined microscopically with the high-power objective. Motile organisms would be seen swimming around.

Plate Count Pr Ml Of Micro-Organisms From The Three Differnet Feed Samples Examined

SAMPLE NO	MEAN TOP FEED	microorganismL VITAL FEED	COUNT PER ML GUINEA FEED
1	100	200	150
2	120	150	180
3	100	150	130
4	100	150	150
5	120	150	150
6	100	150	150
7	100	150	180
8	120	200	130
9	110	200	130
10	100	200	140
11	100	150	180
12	100	150	180
13	100	200	150
14	120	210	150
15	120	210	150
16	100	200	130
17	110	200	180
18	110	200	180
19	110	150	180
20	110	150	180
21	100	150	180
22	100	150	150
23	120	150	150
24	120	150	150
25	100	200	150
26	100	150	150
27	100	200	150
28	100	200	150
29	100	200	180
30	100	200	180
31	100	200	130

32	100	200	150
33	120		
34	120		
35	100		
36	100		
37	110		
38	110		
39	120		
40	100		
41			
42			
43			
44			
45			
46			
47			
48			
49			
50			
AVERAGE	**105**	**166**	**153**

The average plate count per ml of viable microorganisms was highest in vital feed with average count of 166, followed by Guinea feed with average count of 153 and top feed had the least average count of 104.

Table 2
Standard Plate Count Of Different Colonial Forms The Three Different Feed Sampes. Designation Of Colonies

NO	A	B	C	D	A	B	C	D	A	B	C	D
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KEY A is yellow on Nutrient agar and golden on macConkey agar
B is creamy oblique
C is creamy white
D is greenish

Table 3

p^H Reading Od Poultry Feed for The Three Different Poultry Feed Companies.

SAMPLENO	TOP FEED	VITAL FEED	GUINEA FEED
1			
2			
3			
4			
5			
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7			
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50			

The pH values of the feed samples were highest in vital feed with 7.80, followed by Guinea feed with pH 6.46 and top feed had the lowest pH of 6.00

Table 4
Preliminary Identification Of microorganismL Isolates

CODE NO	COLONIAL MORHOLOFY AFTER 24 HOURS AT 37^0C	GRAM REACTION	RESUMPTIVE IDENTIFICATION
A	Yellow discrete convex colonies of about 1-3mm	+	staphylococcus
B	Creamy opaque convex colonies about 2mm with smooth edges	-	salmonella
C	Creamy white spherical colonies of about 1mm with entire edges	+	Bacillus
D	Mucoid confluent growth with greenish pigment of about 1-2mm	-	Pseudomonas

KEY
A is yellow opaque (cocci)
B is creamy opaque (Rod)
C is creamy white (Rod)
D is greenish (Rod)

Table 5
Biochemical Tests for The Identiofication Of Microorganisml Isolates for The 3 Different Poultry Feed Companies.

IDENTIFIED ORGANISM	GLUCOSE	LACTOSE	SUCROSE	H_2S	MOTILITY	NDOLE	CITRATE	CATALASE	VP	MR	OXIDASE
Staphylococcus aureus											
Salmonella typhi											
Bacillus cereus											
Pseudomonas aeruginosa											

KEY
A is acid
G is gas
- is Negative
+ is positive

The types of microorganism isolated from the Poultry feed samples were shown in Tables 5 which included staphylococcus aureus, salmonella typi, Bacillus cereus and Pseudomonas aeruginosa. From the biochemical test above, it was detected that staphylococcus and Bacillus were Acid and gas positive while salmonella and Pseudomonas were only Acid positive in glucose test, while in lactose test all were acid positive except salmonella which was acid and gas positive and in sucrose test, staphylococcus and Pseudomonas was acid positive and salmonella was acid and gas while there was no reaction on Bacillus.

Table 6
Percentage Distribution of Each Microorganisml Isolates from All the Three Different Poultry Feed Companies.

ORGANISMS	AVERAGE COUNT OF EACH ORGANISM	PERCENTAGE DISTRIBUTION

Staphylococcus aureus	52	33
Salmonella typhi	48	30
Bacillus cereus	40	25
Pseudomonas aeruginosa	18	12
Total	158	100

Table 6 shows the percentage distribution or occurrence of the microorganism isolated from the Poultry feed types. Staphylococcus aureus had the highest count of 52 (33%), followed by Salemonellatyphi 48(30%) next was Bacillus cereus 40(25%) and least average count was from Pseudomonas areruginosa 18(12%).

TABLE 7
Average P^{h}reading, Average Count Per Ml And Types Or Microorganisml Organisms Isolated From The Three Different Poultry Feed Companies

TYPE OF FEED	NAME OF COMPANY	AVERAGE pH OF EACH	AVERAGE VIABLE COUNT	TYPES OF microorganismL ORGNAISMS ISOLATED
Starter's mash	Top feed	6.00	105	Staphylococcus aureus Bacillus cereus salmonella typhi
Starter's Marsh	Vital feed	7.80	166	Staphylococcus aureus salemonellatyphi pseudomonas aeruginosa
Starter's marsh	Guinea feed	6.46	153	Staphylococcus aureus salmonella typhi Bacillus seveus

Conclusion

The microorganisml counts obtained in this study were high and indicated the need for improve sanitary conditions in the factories and farms. Much of the contamination may have occurred after processing since the processing techniques utilized did not destroyed many of the micro-organisms, especially the spore forming organisms. Care should be taken during handling and packaging of the finished product and adequate sanitation of Poultry farms will improve the bacteriological characteristics of Poultry feeds.

Recommendations

1. Condition of feed storage and handling could be a source of contamination, therefore hazard Analytical critical control point (Haccp) should be observed during the harvesting of the raw materials, processing, packaging and storage of the finished product.
2. Storage conditions of the feeds should not be in a humid environment

3. To maintain healthy birds, trained personnel should be employed to man Poultry houses to ensure that only properly processed feeds are given to the Poultry birds.
4. Well trained personnels should be employed to work in Poultry feed factories to ensure well processed Poultry feed.

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