



Bacteriological and Physiochemical Screening of Commonly Used Commercial Poultry Feed

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Article Received: August 2020 Published: November 2020

Abstract

Microbial, proximate analysis and heavy metals level were evaluated in some selected commercially available poultry feed samples used as starter, grower, layer and finisher obtained from Port Harcourt Metropolis. Five brands of poultry feeds were evaluated for total heterotrophic count (THC) and presence and absence of *Listeria* and *Salmonella* spp. THC of starter feed ranged from log cfu/g 5.89 -6.98, grower feed 5.29- 6.42, layer feed 5.04-6.07, finisher feed 5.14-6.07 were observed. Organisms isolated include *Bacillus cereus*, *E coli*, *Proteus sp*, *Salmonella spp*, *Pseudomonas spp*, *Micrococcus spp*, and *Enterobacter spp*. Super and hybrid feeds brand had higher THC than the other brands of feeds. Frequency of 3% and 2.5 % occurrence for *Salmonella* and *Listeria* spp, was observed in the poultry feed. The protein content in the brands of feed ranged from 7.34-33.43%, fat ranged from 2.89-7.81%, moisture 3.8-12.2%, ash 6.18-18.21%, carbohydrate 17.19%-61.5% and 3.69-15.6% crude fibre. Zinc, Iron, Copper, Lead, Nickel were found in all the feed in permissible amounts as stipulated by FAO/WHO. The occurrence of these organisms in the feed samples calls for necessary action in the storage methods employed by the poultry and other livestock farmers, the warehouse condition, distributors and the seller. The proximate composition of the different brands of feeds represent great variations among the quality of the poultry feeds from selected manufacturers. A much needed measures should be taken in order to eliminate/reduce the heavy metals from gain access into the feeds thereby reducing human exposure through feeding and there should be provision by Standard Organization Nigeria to provide maximum acceptable limit for heavy metals.

Keywords: *Bacterial, feeds, farmers, warehouse*

INTRODUCTION

Poultry feeds materials that are used to meet the nutritional needs of birds (Obi and Ozugbo, 2007). The materials include grains for carbohydrate, peanuts, fish or bone meals/ for protein . There are different feed types in the Nigerian market. The type of bird the stage of their development and the purpose they are to serve either as meat or for egg, would determine feed selection by the grower. Poultry feed have being implicated in several poultry diseases with varied pathological manifestations. These diseases are of viral (e.g. Avian influenza, Newcastle disease), bacterial (e.g. Salmonellosis) and fungal origin. Feed can act as a carrier for pathogens and aflatoxins due to storage conditions (Maciorowski et al., 2006).

Pathogenic bacteria from the intestinal environment can enter animal feeds. Unattached bacteria or bacteria that are sloughed off with mucosal cells leave the intestinal environment and mix with soil bacteria. A part of this population must then survive in the relatively desiccated and nutrient poor environment until it may colonize another host. If the surviving bacteria is commensal inhabitant of the gut, such as nonpathogenic *E. coli*, their contribution to feed microflora may be of marginal concern. *Listeria* spp. are found in silage and can cause eye infections in ruminant animals (Nightingale et al., 2004). *Listeria* spp., can survive for a long time in some food and go on to cause infection (Aureli et al., 2000). Crump et al. (2002) established that *Salmonella* spp. isolated from poultry is traceable to feed consumed. Feeds made from these animal products such as bone, meat, and fish meal can transmit the *Salmonella* pathogen (Juven et al., 2004)

MATERIALS AND METHODS

Collection of samples

Five different brands of feeds were obtained from the market comprising of the starter, grower, layer and finisher. A total of 20 feed samples were analysed

TOTAL VIABLE COUNTS OF THE COMMERCIAL POULTRY FEED

SAMPLES

225 ml of 0.1% buffered peptone water was transferred into a plastic bag containing 25 g of the poultry feed sample and a homogenized suspension was made. Dilutions ranging from 10¹ - 10⁻¹³ were prepared there from following the recommendation of International Organization for Standardization, 1995.

ISOLATION OF SALMONELLA SPP IN COMMERCIAL POULTRY FEED

25g of feed samples were transferred into 225ml buffered peptone water for pre-enrichment at 37C for 24-48 hours. After which a ml of culture was transferred to 10 ml of selenite F broth and incubation at 370C for 18 hours before plating on Salmonella shigella agar for 24 hours. After incubation period colonies of Salmonella spp colonies were picked from the different plates based on different colonial characteristics and sub cultured onto nutrient agar date for purification before transferring onto Nutrient agar slants and incubated at 370c for 24 hrs. The isolates were characterized presumptively by colonial morphology, Motility, pigmentation, Gram staining and biochemical test including Urease, Sugar fermentation, Indole, Catalase, Methyl-red, Coagulase Test, test, Voges – Proskauer and Oxidase test.

ISOLATION OF LISTERIA SPP IN COMMERCIAL POULTRY FEEDS

Prepared samples were transferred into 225ml half fraser enrichment at 0c for 24 hrs. After which 1ml of the culture was transferred to 10 ml of Full fraser broth and incubated at 370C for 24 -48 hrs. before plating onto PALCAM agar and supplemented with PALCAM Selective Supplement and incubated at 37°C for 24–48 h. After incubation at 370C for 48 hrs, Colonies that appeared grayish colonies surrounded by black halos and sunken centers with possible greenish sheen on PALCAM agar at five characteristic colonies was selected from the Palcam plates and streaked onto tryptone soya yeast glucose agar plates for purification. Isolates were tested for catalase, Gram reaction, motility test, carbohydrate utilization.

PROXIMATE ANALYSIS OF THE VARIOUS SAMPLES

The moisture, protein, fat, ash and carbohydrates contents were determined according to AOAC method (AOAC, 1990). (Bukar and Saeed, 2015).

DETERMINATION OF HEAVY METAL LEVELS IN POULTRY FEEDS

Collection of samples: (25 of each samples) The samples was wrapped in polyethylene bags, and transferred to the laboratory. The samples will be kept frozen until analysis. The samples was analyzed to estimate Cd, Ni, Pb, Cu, Co, Zn, Fe and residual levels by atomic absorption spectrometry (AAS).

Chemicals and reagents

Nitric acid (HNO₃) 65%, Perchloric acid (HClO₄) Hydrogen peroxide (H₂O₂) 70-72%, Hydrochloric acid (HCl) 32% and). All chemicals and reagents used were of the highest purity (Analytical grade). Pure certified atomic absorption standards for lead, cadmium, copper, cobalt and arsenic. Glassware and polyethylene containers were soaked in water and soap for

2 hours rinsed several times with tap water and then distilled water, before using the cleaning mixture solution and finally air dried in incubator following washing with distilled water.

Digestion of samples: Flame atomic absorption spectrometer (AAS) in Perkin Elmer model (spectra-AA10, USA) was used. One gram of chicken meat, feed and egg sample was mashed by sharp scalpel in a screw capped tube. Five milliliters of the acid digestion mixture (3 ml HNO₃: 2 ml HClO₄) was added to the tissue sample. Tubes used throughout were firmly closed and the contents were vigorously shaken and allowed to stand overnight at room temperature. Then, these tubes were heated for 3 hours in water bath adjusted at 70 °C to ensure total digestion of the samples. The digestion tubes were vigorously shaken at 30 minutes intervals. The tubes were then cooled at room temperature and then diluted with 20 ml de-ionized water, and filtered by means of filter paper (Whaitman No. 42). The filtrate was collected in Pyrex glass test tube. The tubes were capped with polyethylene films and kept at room temperature before analysis for heavy metal content. Blanks and standards were prepared in the same manner as for wet digestion and using the same chemicals.

Analysis: The digest, blank and standard solutions were aspirated by the atomic absorption spectrophotometer (AAS) and analyzed for heavy metal contents by air / acetylene flow (5.5 /1.11/m) flame AAS (Buck Scientific Model 210 VGP).

RESULTS

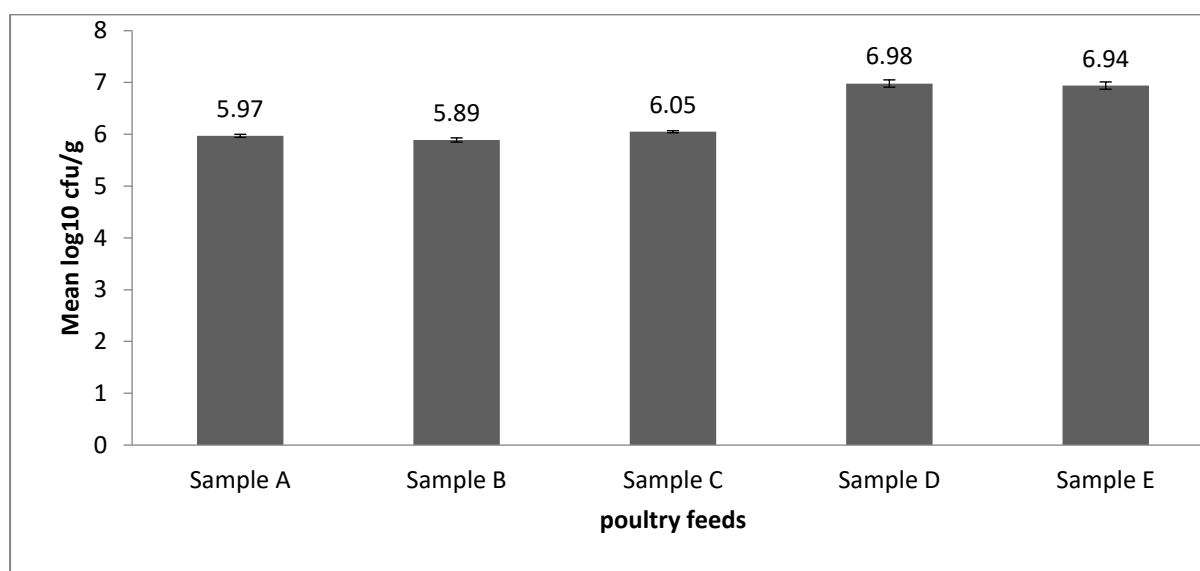


Fig 1 Total Bacterial Counts of The Different Brands Of Starter Feeds

Legend; Sample A- Starter feed ,Sample B- Starter feed, Sample C; Stater feed, Sample D Starer feed ,Sample E – starter feed

Each error bar rep mean \pm std dev

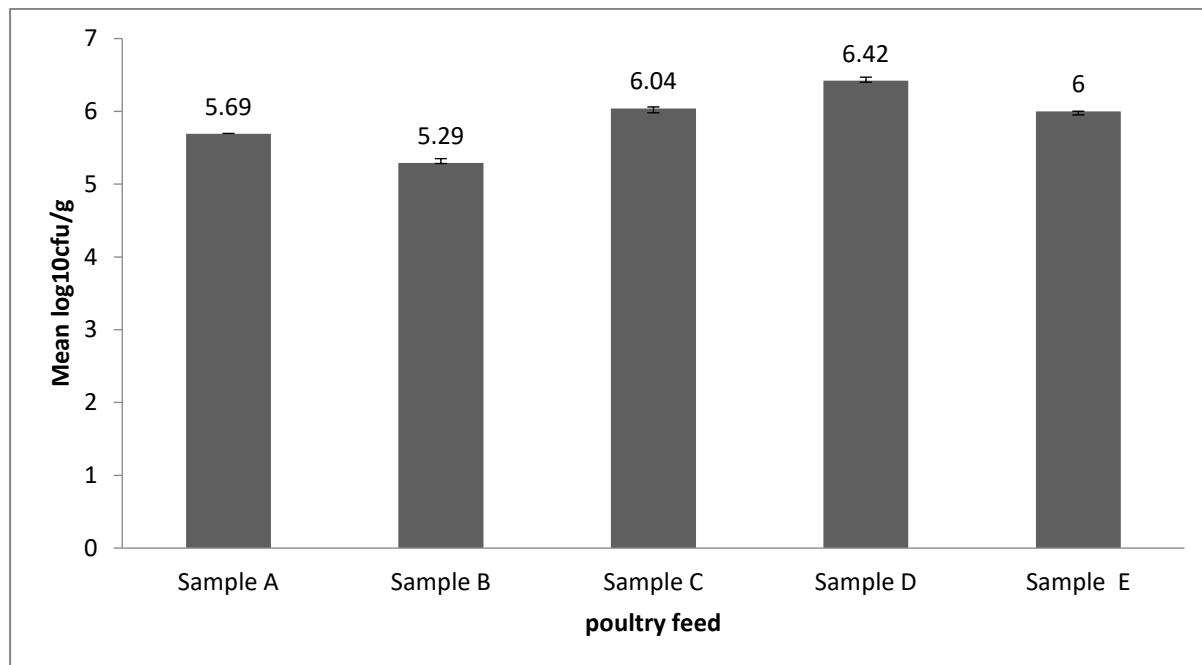


Fig 2 Total Bacterial Counts of the Different Brands Of Grower Feeds

Legend;Sample A- Grower feed ,Sample B- Grower feed, Sample C; Grower feed, Sample D Grower feed ,Sample E – Grower feed

Each error bar rep mean \pm std dev

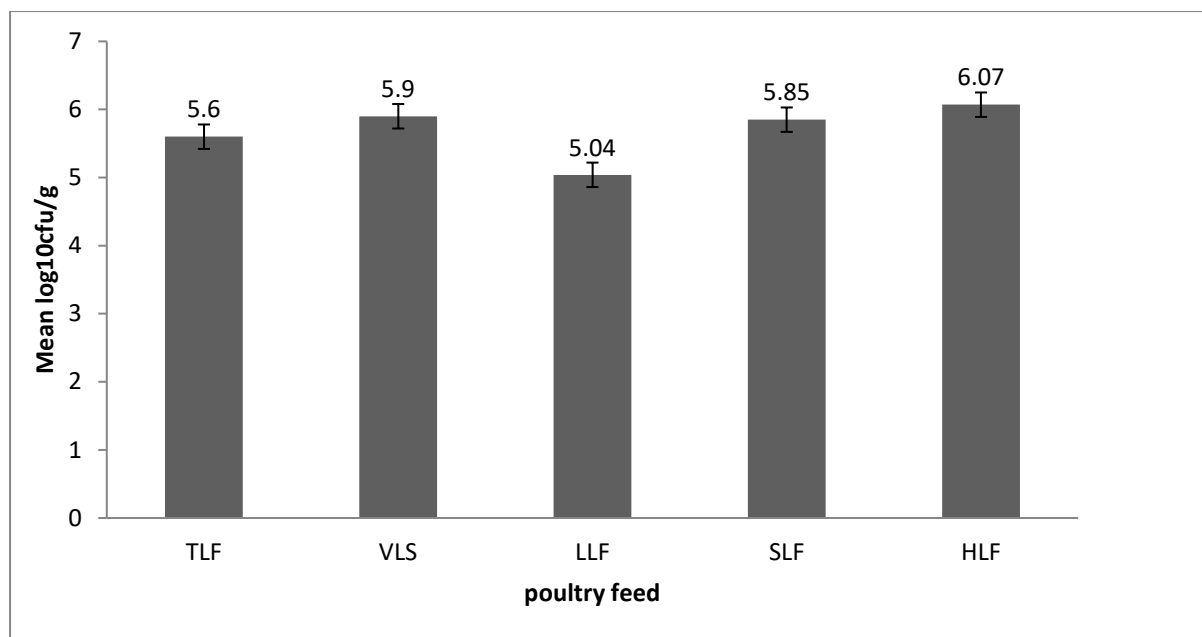


Fig 3 Total Bacterial Counts of The Different Brands Of Layer Feeds

Legend; Sample A- layer feed ,Sample B- layer feed, Sample C; layer feed, Sample D layer feed ,Sample E – layer feed

Each error bar rep mean ± std dev

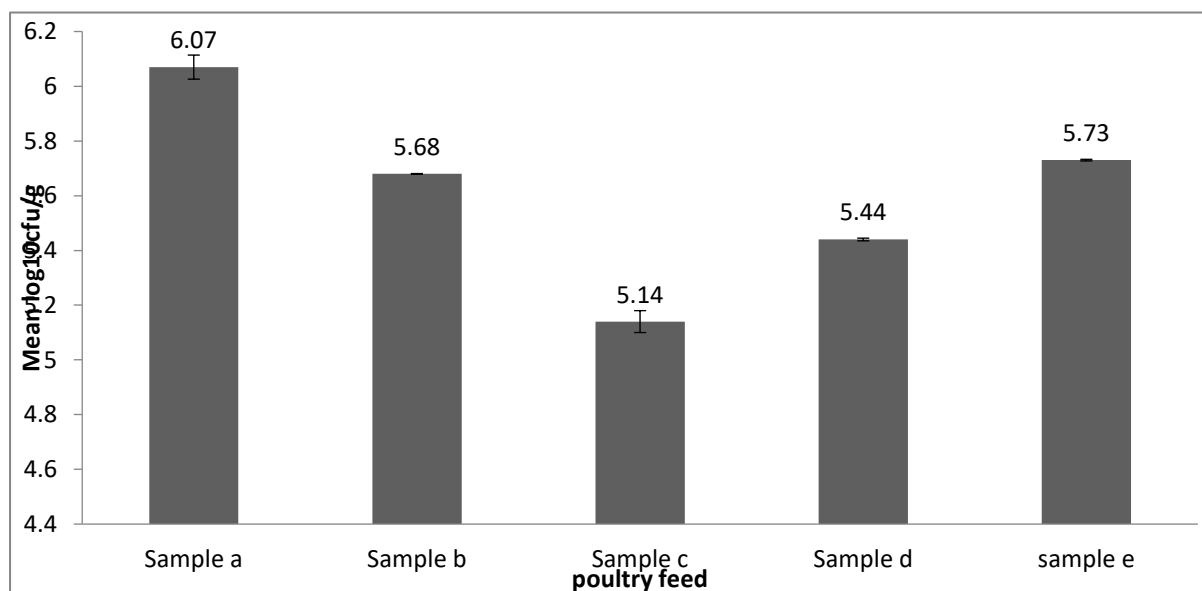


Fig 4 Total Bacterial Counts of the Different Brands of Finisher Feeds

Legend; Sample A- finisher feed ,Sample B- finisher feed, Sample C; finisher feed, Sample D finisher feed ,Sample E – finisher feed

Each error bar rep mean ± std dev

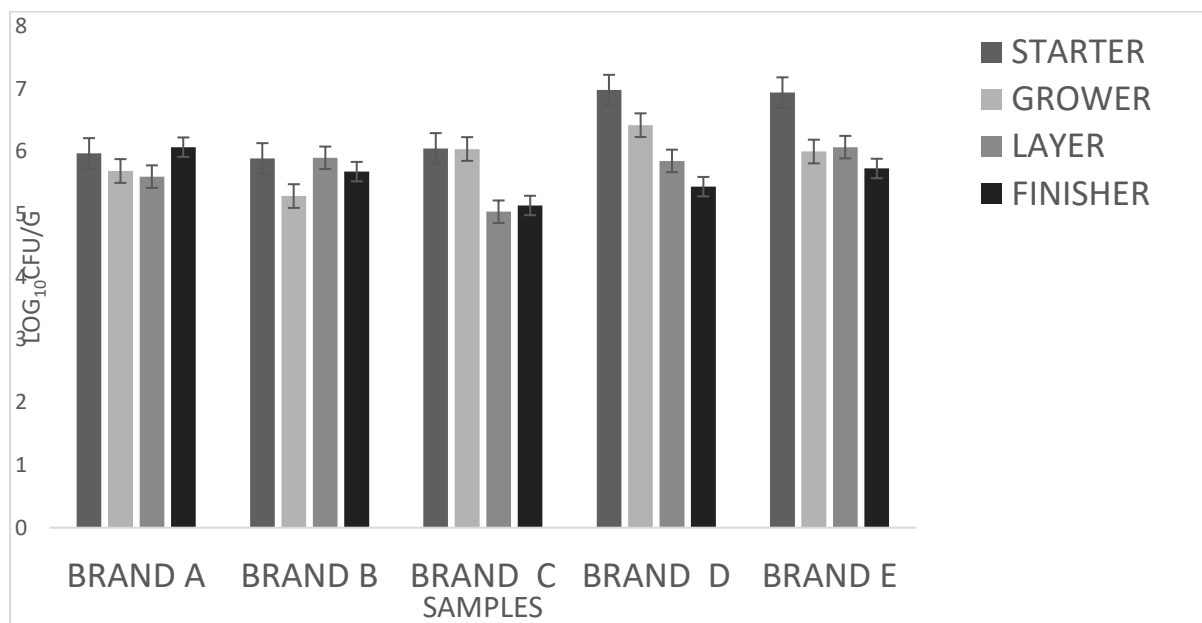


Fig 6 Total Bacterial Counts of The Different Brands of Feeds.

Brand A; Top Feed, Brand B; Vital Feed, Brand C; Livestock Feed, Brand D; Super Feed, Brand E; Hybrid Feed

Each Error Bar Rep Mean ± Std Dev

TYPES OF FEEDS	PRESENCE/ABSENCE OF <i>LISTERIA SPP</i>	PRESENCE/ABSENCE OF <i>SALMONELLA SPP</i>
SAMPLE A FEED		
STARTER	—	—
GROWER	—	—
LAYER	—	—
FINISHER	—	—
SAMPLE B FEED		
STARTER	—	—
GROWER	—	—
LAYER	—	—
FINISHER	—	—
SAMPLE C FEED		
STARTER	—	—

GROWER	—	—
LAYER	—	-
FINISHER	-	—
SAMPLE D FEED		—
STARTER	—	-
GROWER	—	—
LAYER	—	—
FINISHER	—	—
SAMPLE E FEED		—
STARTER	—	—
GROWER	—	—
LAYER	—	—
FINISHER	—	—

Table 1;PRESENCE/ABSENCE OF LISTERIA AND SALMONELLA SPP FROM FEED SAMPLES

Table 2;PROXIMATE COMPOSTION OF THE DIFFERENT BRANDS OF FEEDS

BRANDS						
	CHO	PROTEIN	LIPID	ASH	FIBRE	MOISTURE%
Starter A	26.15	17.65	7.81	14.32	10.14	4.15
StarterB	17.09	15.07	6.08	12.03	5.43	4.53
Starter C	22.43	22.43	3.78	10.76	4.56	5.94
StarterD	17.91	18.04	6.52	6.18	3.67	8.04
Starter E	21.52	21.34	5.78	17.12	6.23	5.67
GrowerA	64.29	10.45	7.06	7.12	4.53	3.8
Grower B	23.18	33.45	8.15	15.76	12.45	7.45
GrowerC	52.73	18.56	7.67	10.15	5.87	8.09
GrowerD	18.64	16.53	2.89	15.58	6.54	12.18
Grower E	18.43	27.3	6.03	18.02	3.87	7.9
LayerA	52.87	11.08	2.9	12.54	15.6	6.9

LayerB	20.63	34.78	6.64	16.84	7.59	7.19
LayerC	40.53	24.31	4.15	14.02	2.08	12.98
LayerD	32.15	23.04	6.89	18.12	4.09	8.15
LayerE	14.71	8.37	5.86	11.21	6.74	11.25
FinisherA	61.15	16.33	6.74	8.75	2.19	4.21
FinisherB	14.89	6.71	7.84	15.78	3.07	7.98
FinisherC	21.73	28.14	4.98	10.12	4	7.01
FinisherD	30.18	23.43	5.62	13.24	3.12	5.78
FinisherE	25.56	7.84	7.43	8.23	4.68	7.16
Mean±SD	29.8±15.7	19.2±8.1	6.04±1.6	12.8±3.7	5.8±3.4	7.3±2.5
Mean±SE	29.8± 3.5	19.2± 1.8	6.04±0.3	12.8±0.8	5.8±0.7	7.3±0.6

Table 3; HEAVY METAL LEVELS IN THE DIFFERENT BRANDS OF FEEDS,

BRAND	A(mg/kg)	ZINC	IRON	COPPER	LEAD	CADIUM	NICKEL	CHROMIUM
Starter		29.1	12.45	2.251	0.007	1.982	1.234	1.121
Grower		32.14	7.15	1.914	1.021	1.621	1.411	0.791
Layer		39.42	9.48	1.412	0.001	1.112	1.112	1.214
Finisher		17.45	11.71	1.721	0.004	1.422	1.221	1.291
BRAND B								
Starter		30.42	18.41	3.132	0.031	2.135	1.112	0.041
Grower		34.25	17.32	4.512	1.015	2.151	1.118	1.116
Layer		22.14	17.14	4.518	1.411	2.113	1.32	2.012
Finisher		34.15	10.41	2.471	0.402	3.017	1.001	1.015
BRAND C								
Starter		31.049	15.32	4.541	1.725	2.321	1.213	1.312
Grower		25.141	20.148	4.821	1.011	2.153	1.009	2.019
Layer		27.421	15.144	5.431	1.015	2.512	1.031	1.252

Finisher	15.142	16.141	4.123	0.312	3.003	1.025	1.214
BRANDD							
Starter	42.321	10.12	2.321	0.012	1.117	2.012	0.151
Grower	31.72	20.43	2.031	0.173	2.013	1.631	1.311
Layer	20.42	12.143	1.432	0.142	3.123	2.031	1.521
Finisher	20.15	20.142	3.115	0.115	1.932	2.134	1.631
BRANDE							
Starter	18.17	20.14	14.31	0.001	1.731	1.312	1.071
Grower	29.43	18.33	2.141	0.004	2.003	2.012	1.301
Layer	28.73	10.32	3.711	ND	1.632	1.763	1.321
Finisher	18.42	21.334	3.321	ND	1.342	1.421	1.031
Mean±SD	27.5±7.5	15.2±4.4	3.66±2.79	0.466±0.55	2.02±0.58	1.41±0.38	1.18±0.48
Mean±SE	27.5±1.7	15.2±0.9	3.66±0.62	0.466±0.12	2.02±0.13	1.41±0.08	1.18±0.1

Discussion

Five brands of poultry feeds were evaluated comprising the starter feed with mean log cfu/g of 5.89-6.98, grower feed 5.29-6.42, layer feed 5.04-6.07, finisher feed 5.14-6.07 respectively. The high bacteria count obtained in the different brands of feed when compared with the international microbiological standard, if it exceeds 300,000cfu/g for older animals and 500,00n for young ones (Anom 2008) it was observed that all the examined feeds were of poor sanitary quality and fail met the standard. This collaborates to the study of Lateef and Gneum-Kana 2014, Omojasola and Kayode(2015)

Many of these organisms isolated represent common environmental contaminants and their presence may indicate contamination from the environment and raw materials during processing. The source of these organisms differ extensively. The bacterial genera maybe from nitrogenous waste products used in compounding animal feeds such as dung, chicken excreta etc as reported by Ogbulie (1995), Organisms isolated include Bacillus cereus, E coli, Proteus sp, Pseudomonas spp, Micrococcus spp, and Enterobacter spp. Animal feeds contaminated with Salmonella could cause infection to livestock and therefore to the human food chain (Crump et al., 2002; Rosa et al., 2005; De Reu, 2006, Krnjaja et al., 2008). Salmonella, Listeria monocytogenes and Clostridium were not isolated from any of the feed samples in this study. The occurrence of these bacteria in the feed samples calls for attention in processing and

storage by the feed manufacturers, distributors and sellers, this is in accordance with the findings of (Chowdhuri et al., 2011).

Poultry farmers must ensure proper disposal of poultry droppings and contaminated feed, to avoid transmissions of pathogen. Presence of *E. coli* indicates faecal contamination while *Staphylococcus*, *Pseudomonas* and *Proteus* spp indicates environmental contamination (Jawetz et al., 1995). Mallinson (1984) opined that *Salmonella* and *Listeria* are pathogens to most birds types. It is very pertinent that producers of poultry feed should create environment and conditions that minimizes feeds contamination. In poultry feeds crude protein, moisture, crude fat, and crude fibre are crucial nutritional compounds (Bukar and Saeed, 2015). The protein content in the brands of feed ranged from 7.34-33.43%, fat ranged from 2.89-7.81%, moisture 3.8-12.2%, ash 6.18-18.21%, carbohydrate 17.19%-61.5% and 3.69-15.6% crude fibre. The data obtained in this study showed great difference in the quality of the feed from selected manufacturers but the constitutive nutrients are basic for sustenance of all bird types.

Zinc, Iron, Copper, Lead, Nickel were found in all the feed in permissible amounts as stipulated by FAO/WHO (2000). Okoye et al, (2012) and Barker and Saeed (2015), observed that levels of these metals in feeds sold in the Nigerian market were within range. Contrary to our study, Mahesar et al. (2010) reported higher levels of these metals in feeds. Cadmium was also found in all the feed samples as was also reported by Bukar and Saeed (2015). Several studies showed that heavy metal such as Nickel, Copper, Zinc, Cadmium, Lead, Chromium in feed were 100 times more than required value (Nakissa et al., 2005).

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Cite this article:

Omorodion Nnenna and Odu Ngozi, "Bacteriological and Physiochemical Screening of Commonly Used Commercial Poultry Feed," *Journal of Multidimensional Research and Review (JMRR)*, Vol.1, Iss.3, pp.06-18, 2020