

ISOLATION AND IDENTIFICATION OF BACTERIAL ISOLATES FROM POULTRY FEED CONTAMINATIONS AT PUDUKKOTTAI DISTRICT, TAMIL NADU.

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Abstract

Poultry feed can serve as a carrier for a range of microbial contaminants such as moulds, mycotoxins and bacteria. This study was done to determine isolates from three different brands of broiler poultry feeds. Organisms isolated from poultry feeds were *Staphylococcus aureus*, *E.coli*, *Bacillus subtilis*, *Salmonella* sp, *Proteus* sp and *Shigella* sp. Among the bacterial isolates of all samples, Gram negative bacteria were common all the samples especially *E.Coli* with 40%. The presence of some pathogenic microorganisms in the poultry feeds revealed high level of contaminations. It is recommended that poultry feeds should be made from good quality grains and it should

Poultry feeds are food materials used in raising poultry birds. The term 'poultry' used in Agriculture generally refers to all domesticated birds kept for egg laying or meat production. It is the second most eaten meat in the world for about 38% (Raloff, 2003).It is referred to as complete feed as they are designed to contain all the

be prevented from environmental or other contamination.

Keywords: Poultry feeds, bacterial contaminants, *E.coli*, pathogenic bacteria, *Proteus* sp.

1. Introduction

Feeds are good and nourishing food supplements with varying constituents of animal and vegetable proteins, cereals, essential amino acids, minerals, salts, antibiotics, amino acids etc., Since commercial feed and feed ingredients are usually sourced from various locations, they remain the major vehicles for the introduction of both commensal and pathogenic microbes to the farm environment (Okala *et al.*, 2005). nutritional materials needed for their proper growth. The diseases of poultry are like that of the disease of other animals. They may be caused by pathogenic organisms, nutritional deficiency and from wound or cannibalism. Some of the diseases associated with fowls locally include; new castle disease, chronic

respiratory disease, fowl typhoid and fowl pox diseases (Onyene *et al.*, 2013).

To prevent pathogenic organisms from getting into the body of poultry, attention should be given to the factors that influence their infectious spread. First and foremost, they should have disease spreading stock, clean range, proper feeding

2. Materials and Methods

2.1 Collection of Samples

Poultry feed samples from various broiler farm were collected in a sterile polythene bags in Pudukkottai District, Tamil Nadu. Then the three collected feed samples were labeled and taken to laboratory for microbiological analysis.

2.2 Isolation of bacteria from collected samples

Feed samples (1g each) were taken separately into nutrient broth media (Hi-Media, India), and incubated at 37 °C for 48hrs aerobically. After incubation one loopful of each suspension was separately inoculated into nutrient broth media. Control flasks without inocula were also prepared and incubated at 37 °C. Then the broth was subculture into Nutrient agar. All these plates were incubated at 37 °C for 48hrs aerobically.

and quarantining new stock. Sanitation is very important in poultry management by cleaning of their water can, feeding troughs and finally disinfecting to help reduce organic matter. The aim of this research was to isolate and determine the contaminants that are associated with poultry feeds in Pudukkottai District, Tamil Nadu.

2.3 Identification of bacterial isolates

The bacterial isolates were characterized and identified by observing their gross colony morphology grown in Nutrient agar, EMB, MacConkey, Salmonella-Shigella (SS), XLD agar. The identities of the microbes were confirmed employing an approved technique (Buchanan *et al.*, 1985). In addition, the isolates were subjected to conventional Gram staining, motility test, biochemical tests. Such as indole productivity, H₂S, Simon citrate, catalase, coagulase, oxidase and sugar utilization, following the standard methods (Cowan, 1985).

3. Result

This study was conducted in order to determine and investigate the bacterial contaminants of broiler poultry feeds. Three different poultry feed samples were examined and a total of 384 bacterial

isolates were recorded in Table.1 Organisms isolated from three various brand poultry feed samples are *Staphylococcus aureus*, *E.coli*, *Bacillus subtilis*, *Salmonella* spp, *Proteus* sp and *Shigella* sp. Among the all bacterial isolates *E.coli* were the common

bacterial contaminant found in the poultry feed collected from Pudukkottai District, Tamil Nadu were identified with morphological and biochemical test represented in Table.2 and 3.

Table 1: Total plate count of the Bacterial count from Poultry feed samples

Samples from different brands	Plate Count
PS-1	132
PS-2	141
PS-3	111
Average	384

Key: PS indicates Pudukkottai Sample

Table 2: Cultural and Biochemical Characteristics

Cultural Colonies Character ics	H ₂ S	Glucose	Lactose	Acid	Gas	Motility	Indole	Citrate	Coagulase	Catalase	Oxidase	Gram Staining	Shape	Organism
Creamy and convex with smooth edges	-	+	+	+	+	+	+	-	+	+	-	-	C	<i>Staph.aureus</i>
Pink, convex with smooth edges	+	+	+	+	+	+	+	-	-	+	-	+	R	<i>E.coli</i>
Flat and white on agar plate	-	+	+	-	-	-	-	-	-	+	-	+	R	<i>Bacillus subtilis</i>
Creamy, round, convex smooth edges	-	+	+	+	+	+	-	+	-	+	-	-	R	<i>Salmonella</i> sp
Colonies were white and swampy	-	+	+	+	+	-	+	-	-	+	-	-	R	<i>Proteus</i>
Black convex colonies with smooth edges	+	+	+	+	+	-	-	-	-	+	-	+	R	<i>Shigella</i> sp

Key: + indicates positive, - indicates negative, C indicates Cocci, R indicates Rod

Table 3: Bacteria isolated from Poultry feed samples

Samples	Bacterial Isolates
Sample-1	<i>Staphylococcus aureus</i> <i>E.coli</i> <i>Salmonella</i> sp
Sample-2	<i>Bacillus subtilis</i> <i>Salmonella</i> sp <i>E.coli</i>
Sample-3	<i>Proteus</i> <i>Shigella</i> sp <i>E.coli</i>

Table 4: Standard plate count of different bacterial isolates

No. of Samples	A	B	C	D	E	F
Sample-1	40	52	40	-	-	-
Sample-2	-	62	28	51	-	-
Sample-3	-	40	-	-	28	43
Average	40	154	68	51	28	43

Key: A- *Staph.aureus*, B- *E.coli*, C-*Salmonella* sp, D-*Bacillus subtilis*, E-*Shigella*, F- *Proteus* sp

Table 5: Percentage distributed of Bacteria isolated

Bacterial isolates	Mean plate count	Percentage Distribution
<i>Staphylococcus aureus</i>	40	10%
<i>E.coli</i>	154	40%
<i>Salmonella</i> sp	68	18%
<i>Proteus</i>	43	12%
<i>Shigella</i> sp	28	7%
<i>Bacillus subtilis</i>	51	13%

Table.3 represents the total plate count of bacteria for all of the samples on the selective media. The counting is based on the colours of the colonies. Finally Table.5 represents the percentage distribution of isolated bacteria from the poultry feed sample. *E. coli* had the highest percentage of 40% distribution, followed by 18% *Salmonella* sp, 13% *Bacillus subtilis*, 10% *Staphylococcus aureus* and finally 7% of *Shigella* sp.

4. Discussion

Animal feed has been listed at once as one of the main source of microbes of microbe in farm animals and poultry. The high occurrence of fungal and bacterial species is of public health concern and this may indicate obvious health hazard in term of direct consumption of contaminated feed or their toxins by farm animals (Aliyu *et al.*, 2016). *E. coli* had the highest percentage of 40% in poultry feed contamination among the other microbes. Similarly However higher prevalence of *E. coli* (71.80%) was observed by Derakhshantar and Ghanbarpour (2002) might be due to different environmental condition, managemental condition, food habit and mixed infection with other microbes.

The bacterial species in the poultry feeds include *Bacillus*, *Escherichia*, *Salmonella*, *Enterococcus*, *Campylobacter*, *Clostridium* and *Lactobacillus* that have been shown to be of critical importance in tropical countries like Bangladesh (Noohi *et al.*,2014). *Salmonella* sp. and *E. coli* were isolated and identified from seven poultry feeds in Dhaka, Bangladesh, in which TVC values were 6.75×10^4 and 3.05×10^4 , respectively (Chowdhuri *et al.*, 2011).

In this study, *E. coli* plays the predominant number among all bacterial isolates. In Nigeria, however, species of pathogenic bacteria such as *Streptococcus*, *Bacillus*, *E. coli*, *Salmonella*, and *Pseudomonas* were isolated from commercially available feeds (Uwaezuoke *et al.*,2008). Bacterial counts in starter, grower, finisher and layer poultry feeds using pour plate technique were studied, where *E. coli* (42.0%), *Salmonella* (24.4%) and *Proteus* (33.6%) were found as the major poultry feed contaminants in Nigeria (Onyeze *et al.*,2013). The present findings are somewhat different from those of the aforesaid ones, both in terms of the bacterial species and their load in poultry feeds perhaps owing to the difference in manufacturing, handling and distribution of the feed items, coupled with differences in

the levels of hygiene, bio-safety measures and management practices in rearing chickens in the study area.

5. Conclusion

From the present study it could be concluded that the commercial broiler farms should be periodically checked for the presence of pathogens and the prevention of microbial infections and biosecurity plan to the farms should be taken accordingly. The absence of pathogenic microorganisms isolated from poultry feeds suggests that the food processing is well handled. In recent years, with the increasing in density of poultry and infectious diseases in poultry caused by pathogenic bacteria, the healthy development of the poultry industry is facing serious threat.

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