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MICROBIAL CONTAMINANT ISOLATION FROM POULTRY FEEDS AND REMEDY TO OVERCOME IT

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Abstract: Poultry farming is a practice done to raise birds like Chickens, ducks, turkeys, etc. domestically or commercially for meat, eggs, and also for feathers. Nowadays poultry industry is in great demand. To improve the yield and production of poultry by-products it is necessary to maintain the health of poultry birds. The health of poultry birds is depending on the poultry feed and the growing environment the birds. The microbial contaminants present in poultry feed are the biggest source to reduce the health of poultry birds. In this research article, the study has been carried out to isolate the microbial contaminants from different types of commercial and domestic poultry feeds. The study has been carried out to find a remedy to reduce the contaminants. To reduce the contaminant UV radiation treatment was studied. The study has great importance and applications in the future.

Index Terms - **Poultry Feed, UV radiation, Microbial Contaminants, Chickens, Poultry Farming.**

I. INTRODUCTION

Poultry farming is a practice done to raise birds like Chickens, ducks, turkeys, etc. domestically or commercially for meat, eggs, and also for feathers [1]. Chickens, ducks, and turkeys are having primary importance and demands while young pigeons and guinea fowl are depends on local interest [2]. While rising poultry birds for commercial purposes large scale it is necessary to maintain the proper health and growth of the birds [3]. The good health and growth of poultry birds is depending on the appropriate diet for birds [4]. Poultry feed from commercial brands is mostly preferred in large-scale poultry farming [5]. Commercial poultry feedings are made by ensuring the maximum energy and nutrient intake for the growth and production of fat in birds [6]. A high intake of good quality and balanced protein sources can help in the maximum development of organs, muscles, feathers, and skin in poultry birds [7]. Commercial poultry feedings supplemented with essential minerals can help in the development of bones and eggs [8]. As the growth and development of poultry birds majorly depend on the feed, the feed should be made by following the nutritional demands, balanced nutrient quantity, and proper hygiene during the making of poultry feed [9]. Inappropriate hygiene results in contamination in the feed, which ultimately shows effects on the health of the poultry birds [10]. The causes of microbial contamination in feed include contaminated raw materials, supplementary products, animal wastes, human wastes, rat or mouse droppings, contaminated water, etc. [11] The feed may also be microbially contaminated by coming in contact with soil microflora, contaminated air microflora, fecally contaminated components. The microbial contaminants mostly found in poultry feed include *Staphylococcus aureus, E. coli, Bacillus subtilis, Salmonella spp., Proteus mirabilis, Shigella spp., Corynebacterium spp.,* etc. and also includes fungal contaminants [12][13]. Some of these microbial contaminants are the indicators of fecal contamination such as organisms belonging to the group of coliforms [14]. These contaminants can cause serious infections and illness in poultry birds which may result in a reduction of Commerciale poultry products and their yield [15]. The presence of contaminants in the feed may sometimes result in the death of the poultry birds [16]. To overcome this problem, there are various preventive practices should have to be performed. It includes an aseptic collection of raw material, aseptically making of feeds from grains, and aseptically packaging by removing all moisture contain from the feed for long-term storage purposes [17]. Some chemical disinfectants are also used in making contaminant-free poultry feed [18]. Heat treatment and exposure to UV radiation are also effective methods for making contaminant-free poultry feed [19].

II. Material and Methods

2.1 Sampling

A total of 10 samples of different types of poultry feeds were collected from the local markets and poultry farms located near the region Kopargaon, Ahmednagar, Maharashtra, India. The different types of poultry feeds include Chick Starter, Layer, broiler super starter, Broiler Starter, Broiler Finisher, grower's mash, and Household kadaknath feed. Sampling was done aseptically and the samples were stored in sterile airtight containers.

2.2 Screening for Microbial Contaminants

To check the presence of microbial contaminants in the sampled poultry feed, the tenfold dilutions of samples were made individually using the serial dilution method (10g sample in 90ml sterile distilled water) [20]. One drop from each dilution was inoculated on sterile nutrient media plates and the plates were incubated overnight at 37°C. After incubation, those samples which showed growth on nutrient media plates were taken and CFU counts were measured in triplicates of each sample. The poultry feed samples which showed contamination were selected for further analysis [21].

2.3 Microbial Contaminant Isolation and Characterization

For the isolation and characterization of screened contaminants, a loopful culture of diluted samples was inoculated separately on different selective media plates using the streaking method to obtain isolated colonies [22]. Further pure cultures were obtained from isolated colonies grown on different selective media. After getting pure isolates, the morphological and gram characters of each isolate were recorded. further biochemical tests were performed to identify each isolate up to genus level [23]. The biochemical test includes IMViC, Carbohydrate test, and test for enzymes [24][25].

2.4 UV Treatment to Poultry Feed

The poultry feeds of all selected types were taken aseptically in completely dry sterile Petri plates for exposure to UV radiations at the lab scale. The plates were exposed to UV radiations by keeping them for different time laps under the laminar airflow [26].

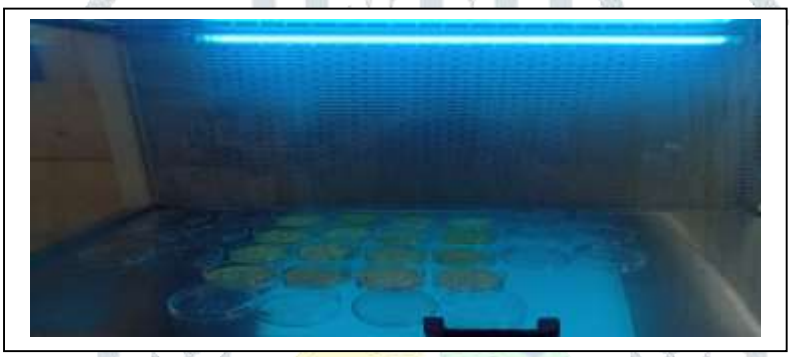

Figure 2.4.1. UV Treatment to Poultry Feed

2.5 Screening for Microbial Contaminants after UV Treatment

After exposure of all the poultry feed samples to UV radiations at different time laps, all the samples of different types and different time laps were again serially diluted following the same process and individually inoculated on sterile nutrient media plates. The inoculated plates were incubated for 24 h at 37°C. The CFU count of each plate of contaminated poultry feed sample was measured after incubation to check the effect of UV radiation on the reduction or removal of microbial contaminants present previously in the selected poultry feeds [27].

III. Result and Discussion

3.1 Screening of Microbial Contaminants

All poultry feed samples were screened for the presence of contaminants on nutrient agar plates. The CFU/ml count of only those plates showing contamination was measured. Which is given in Table 3.1.1

Table 3.1.1. CFU/ml count of colonies present on nutrient media for screening of contaminants in samples

Samples / Plates	Chick Starter	Broiler Starter	Layer	Kadaknath
Replicate-1	38×10^{-5}	55×10^{-5}	33×10^{-5}	29×10^{-5}
Replicate-2	41×10^{-5}	59×10^{-5}	32×10^{-5}	39×10^{-5}
Replicate-3	36×10^{-5}	61×10^{-5}	37×10^{-5}	36×10^{-5}
Average CFU/ml	38×10^{-5}	58×10^{-5}	34×10^{-5}	34×10^{-5}

Among all 10 poultry feed samples, four samples showed the highest contamination which includes Chick Start 38×10^{-5}, Broiler Starter 58×10^{-5}, Layer 34×10^{-5}, Household Kadaknath feed 34×10^{-5}

Figure 3.1.1. Screening of Contaminants on Nutrient Agar Plates

3.2 Morphological and Biochemical Analysis of Isolates

The four selected poultry feed samples which are having high contaminants were further inoculated individually on selective media plates and pure cultures obtained from them were examined for colony characterization and gram characterization. The biochemical characterization of all four samples was also done. The observations are given in Table (3.2.1)

Table 3.2.1. Morphological and Biochemical Characterization of Microbial Isolates from Poultry Feeds.

Sr. No	Test	Chick Starter		Broiler Starter	Layer	Kadaknath	
1	**Media**	MSA	MaC	EMB	MaC	SS	EMB
2	**Gram - character**	Positive	Negative	Negative	Negative	Negative	Negative
3	**Shape and arrangement**	Cocci	Rods	Rods	Rods	Rods	Rods
4	**Cultural characteristics**						
	Size	1-2 mm	1-2 mm	2-3 mm	1-2 mm	1-2 mm	2-3 mm
	Shape	Round	Circular	Circular	Circular	Round	Circular
	Pigmentation	Golden Yellow	NIL	NIL	NIL	NIL	NIL
	Margin	Entire	Entire	Entire	Entire	Entire	Entire
	Elevation	Flat	Convex	Raised	Convex	Convex	Raised
	Opacity	Opaque	Translucent	Opaque	Translucent	Translucent	Opaque
	Consistency	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth
	Motility	Non-Motile	Motile	Motile	Motile	Non-Motile	Motile
5	**Carbohydrates fermentation**						
	Lactose	+ ve	- ve	+ ve	- ve	- ve	+ ve
	Dextrose	+ ve	+ ve	+ ve	+ ve	- ve	+ ve
	Sucrose	+ ve	- ve	+ ve/- ve	- ve	- ve	+ ve/- ve
6	**Biochemical test**						
	Indole Production	- ve	- ve	+ ve	- ve	+ ve/- ve	+ ve
	Methyl Red Test	+ ve	+ ve	+ ve	+ ve	+ ve	+ ve
	Vogus Proskuer Test	+ ve	- ve	- ve	- ve	- ve	- ve
	Citrate Utilization test	+ ve	- ve	- ve	- ve	- ve	- ve
7	**Enzyme**						
	Catalase	+ ve	+ ve	+ ve	+ ve	+ ve	+ ve
	Oxidase	- ve	- ve	- ve	- ve	- ve	- ve
	Amylase	- ve	- ve	- ve	- ve	- ve	- ve

***MSA- Mannitol Salt Agar, EMB- Eosin Methylene Blue, MaC- MacConkey Agar, SS- Salmonella Shigella Agar, + ve- Positive, - ve- Negative.**

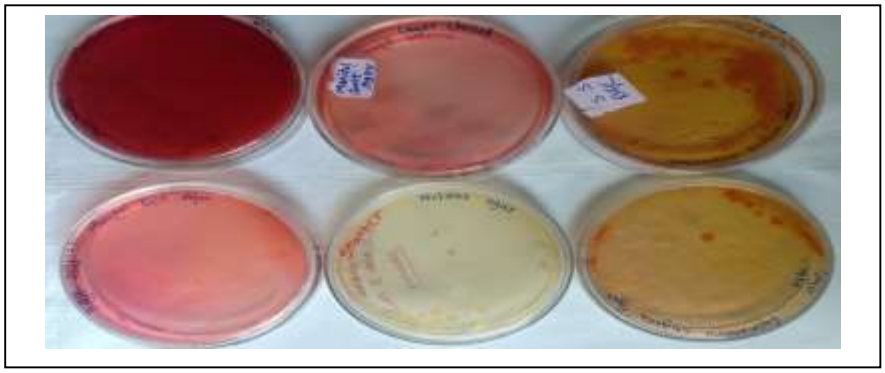

Figure 3.2.1 Growth of Microbial Isolates on Selective Medias

Figure 3.2.2 Observations of IMViC Tests

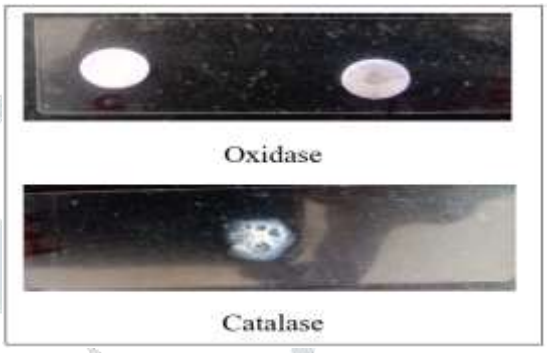

Figure 3.2.3 Observations of enzyme Tests

After analysing the results of morphological and biochemical characterization tests and by comparing the observations given in (Table 3.2.1) with Bergey's Manual of Determinative Bacteriology, it was found that the four microbial contaminants screened and isolated from different poultry feed samples may tentatively belong to *Staphylococcus spp., cc, Shigella spp., Salmonella spp.* In the sample of Chick Starter *Staphylococcus spp.* and *Salmonella spp.* were found. In the sample of Broiler Starter, *E. Coli* was found. In the sample of Layer, *Salmonella spp.* was found and in the sample of domestic kadaknath feed, *Shigella spp.* and *E. Coli* were found. The contaminations in all the studied commercial and domestic poultry feeds may occur due to inappropriate and unhygienic practices during the making and packaging of poultry feeds.

3.3 Screening of Microbial Contaminants after UV Treatment

After giving UV exposure to selected poultry feed samples at different time-lapses, the samples were again tested for the presence of contaminants. The same method of initial screening was repeated and the CFU count was measured, which is given in (Table 3.3.1).

Table 3.3.1 CFU/ml count of colonies present on nutrient media for screening of contaminants in samples after UV radiation treatment.

Samples / Time-lapse	Chick Starter	Broiler Starter	Layer	Kadaknath
10 min	11×10^{-5}	14×10^{-5}	12×10^{-5}	15×10^{-5}
20 min	8×10^{-5}	9×10^{-5}	7×10^{-5}	11×10^{-5}
30 min	5×10^{-5}	6×10^{-5}	5×10^{-5}	7×10^{-5}
40 min	3×10^{-5}	2×10^{-5}	2×10^{-5}	4×10^{-5}

Figure 3.3.1 Screening of Microbial Contaminants after UV Treatment on Nutrient Agar Plates

After screening the samples for the second time by following UV radiation treatment, it was found that the number of colonies present on nutrient media was decreased as compared to the first screening without UV radiation treatment. According to observations and the CFU count is given in (Table 3.3.1) the number of colonies or CFU count decreased with an increase in time of UV radiation exposure.

IV. CONCLUSION

Maintaining hygiene is a very important parameter that should have to be taken into consideration while making human food or animal feeds. Before poultry feed, the presence of the above microbial contaminants in feed is an indication of poor hygiene, lack of cleaning, and fecally contaminated sources or areas near the production and packaging of poultry feed. Such microbial contaminants affect the health of poultry birds which ultimately reduces the production and income of poultry farmers. To overcome this problem so many methods are available. The method used to remove the contaminant from feed should not have any side effects on the health of poultry birds, it should not affect the content and nutritional quality of feed. Considering all these quiescence's effective and convenient method to remove microbial contaminants from poultry feed was found to be the UV radiation treatment to the feed. Before packaging, the prepared poultry feed should be exposed to UV radiation several times and immediately pack.

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