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Evaluation of microbiological and chemical determinations of the raw poultry and the fermented poultry waste manure

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Abstract
During the present study evaluation of microbiological and chemical determinations of the raw poultry and the fermented poultry waste manure was conducted. Highest bacterial count was reported in raw poultry manure with a value of 8.59 the bacterial count was minimum in fish feed with a value of 4.07 while it was 6.50 in fermented poultry manure. The results of the chemical determinations revealed the pH of the raw poultry with higher values of 8.7±0.1 while for fermented poultry manure the pH was 4.0±0.5. The ash level in raw poultry reported was 20.1±1.5. Similarly crude protein content was higher in raw poultry manure with a value of 24.6±2.6 while it was 22.9±3.4 in fermented poultry manure. Non protein nitrogen was lower in raw poultry manure (0.15±0.1) as compared to fermented poultry manure (0.36±0.5).

Keywords: Microbiological, chemical determinations, raw poultry manure, bacterial count and fermented poultry manure

Introduction
Fishes are a rich source of high-quality protein, containing essential amino acids necessary for human health. Consuming fish as part of a balanced diet can contribute to protein requirements. In addition to protein, fishes provide essential nutrients such as omega-3 fatty acids, vitamins (e.g., vitamin D), and minerals (e.g., iodine, selenium), which are important for overall health, brain development, and disease prevention. Fishing and aquaculture provide employment and income for millions of people globally, particularly in developing countries where small-scale fisheries and fish farming are prevalent. The fishery sector contributes significantly to the economies of many countries, both through domestic consumption and international trade (Omojowo and Omojasola, 2013) [4].

Fish pond manuring is indeed a common practice in aquaculture for enhancing fish production and maintaining ecological balance within the pond ecosystem. Manure, typically sourced from livestock or poultry operations, serves as a nutrient input into the fish pond ecosystem. It contains organic matter rich in nitrogen, phosphorus, and other essential nutrients required for plant and algae growth. In fish ponds, the manure is directly consumed by fish, serving as a source of food and nutrients. As the fish feed on the organic matter, they excrete waste, releasing nutrients back into the water. This nutrient recycling process helps to maintain a balanced ecosystem within the pond. The nutrients released from fish excreta and decomposing organic matter support the growth of photosynthetic organisms, such as phytoplankton and aquatic plants. These organisms utilize sunlight to convert carbon dioxide and nutrients into organic matter through photosynthesis (Moav *et al*., 1977, Little and Edwards, 1999) [9, 3].

The preference for chicken manure in agricultural practices, particularly in the context of its ready solubility and high phosphorus concentrations, is well-documented. Poultry waste is rich in nutrients such as nitrogen and phosphorus, which are essential for growth. Poultry waste may contain pathogens such as bacteria, viruses, and parasites that can contaminate water sources if not properly managed. Pathogens present in poultry waste can pose risks to human health if ingested through contaminated water or food crops irrigated with contaminated water. Poultry manure may contain pathogens such as bacteria, viruses, and parasites, which can pose risks to human health and aquatic organisms if washed into water bodies. Pathogens in manure can survive in water environments and may cause waterborne illnesses if ingested or contacted by humans or animals (Sovova, 2012) [6]. The microbiological analyses conducted on manure samples have indicated the presence of various pathogenic microorganisms. Research has shown that zoonotic pathogens, which are capable of infecting both animals and humans, can survive in such environments for extended periods, with some studies reporting survival times of up to 4 months. Factors such as temperature, pH, oxygen levels, and ammonia concentration in the environment can influence the survival of pathogens. For example, pathogens may survive longer in environments with favorable conditions such as moderate temperatures and neutral pH (Jones, 1976, Guan and Holley, 2003) [7, 5]. Keeping this in consideration the present study was conducted to determine microbiological quality of fish feed, raw poultry manure and fermented poultry manure and changes in chemical determinations of the raw poultry and the fermented poultry waste manure.

Materials and Methods

The study conducted at the Laboratory of the Department of Zoology, B. N. Mandal University in Madhepura, Bihar, India. The study was conducted to determine microbiological quality of fish feed, raw poultry manure and fermented poultry manure and changes in physical parameters of the raw poultry and the fermented poultry waste manure.

Microbiological analysis

10 grams of each sample were blended with 90 mL of saline water (0.9% NaCl) using a warring blender. Colony forming units (CFU) were determined using standard pour plate methodology. This involves diluting the sample in a series of dilutions and then plating a known volume of each dilution onto agar plates. After incubation, colonies formed by viable microorganisms are counted, and the CFU per unit volume of the original sample can be calculated. Decimal dilutions of the initial suspension were made using 0.85% saline solution. This dilution series allows for the estimation of total viable counts of microorganisms present in the sample. 1 mL of each dilution was plated in duplicate on standard plate count agar (Biokar, France), a nutrient-rich medium suitable for the growth of a wide range of microorganisms. Enterobacteria, a group of bacteria that includes many pathogens and facultative anaerobes, were specifically enumerated. The enumeration of enterobacteria was performed on MacConkey Agar (Leininger, 1976) [8], a selective and differential medium commonly used for the isolation and enumeration of Enterobacteriaceae. After plating, the plates were incubated at 37 °C for 24 hours, allowing the growth of colonies of enterobacteria. All agar plates were incubated at 37 °C for 24 hours to promote the growth of viable microorganisms. This temperature is conducive to the growth of many mesophilic bacteria commonly found in environmental and food samples.

Chemical determinations

The pH of a solution was measured using a pH meter. Specifically, a Crison Micro-pH 2000 pH meter was used. The dry matter content of a sample was determined by weighing a specific amount of the product and then kept it on hot-air oven drying at 105 °C until its weight no longer changes. This process removes all moisture from the sample, leaving only the dry matter behind. The ash content of a sample was determined by igniting it at a high temperature, specifically 550 °C. The total nitrogen content of a sample was determined using the Kjeldahl method. This method, described by the American Public Health Association (APHA) in 1989. NPN refers to nitrogen in a sample that is not in the form of proteins. To measure NPN, the filtrate obtained after precipitating the proteins with a 10% trichloroacetic acid solution was used and measured using the method followed by Conway (1947) [2].

Results and Discussion

Different microbiological analysis like Total bacterial count was reported in Fish feed, Raw poultry manure and Fermented poultry manure during May 2019 to April 2021. Highest bacterial count was reported in Raw poultry manure with a value of 8.59, the bacterial count was minimum in fish feed with a value of 4.07 while it was 6.50 in Fermented poultry manure (Table 1). Different types of manure, such as poultry, swine, or cattle manure, may harbor different pathogens and have varying levels of organic matter and nutrients, influencing pathogen survival. Pathogen survival is often influenced by temperature, with warmer temperatures generally favoring increased survival rates (Jones, 1976; Guan and Holley, 2003) [7, 5].

Table 1: Estimation of microbiological quality of fish feed, raw poultry manure and fermented poultry manure

Months	X10cfu/ml		
	Fish feed	**Raw poultry manure**	**Fermented poultry manure**
	TBC (Total bacterial count)	**TBC (Total bacterial count)**	**TBC (Total bacterial count)**
May19	4.0	8.2	6.0
Aug19	4.0	8.2	6.2
Nov 19	4.2	8.4	6.4
Feb20	4.0	8.4	6.6
May20	4.0	8.6	6.6
Aug 20	4.4	8.4	6.6
Nov20	4.0	8.5	6.5
April21	4.0	8.6	6.5
Average	4.07	8.59	6.50
SD	0.24	1.2	0.76

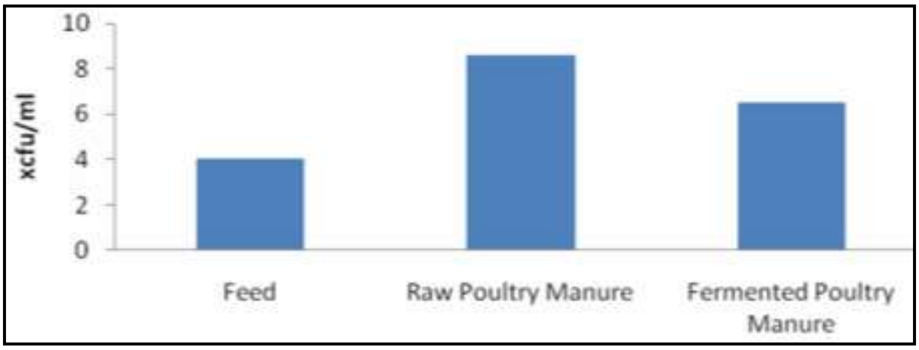

Fig 1: Estimation of feed, raw poultry manure and fermented manure

In the present study the pH of the raw poultry reported was 8.7±0.1 while for fermented poultry manure the pH was 4.0±0.5. In raw poultry Dry Matter reported was 79.3±3.5 for fermented poultry manure. The ash level in raw poultry reported was 20.1±1.5 and fermented poultry manure it was 20.2±3.5 (Table 2). Similarly crude protein content was higher in raw poultry manure with a value of 24.6±2.6 while it was 22.9±3.4 in fermented poultry manure. Non protein nitrogen was lower in raw poultry manure (0.15±0.1) as compared to fermented poultry manure (0.36±0.5). Total volatile Nitrogen proteins were found absent in fermented poultry manure while a level of 4.9±0.2 was reported in raw poultry manure. There were elevated levels of reducing sugars in fermented poultry manure (4.5±2.1) as compared to raw poultry manure (1.2±0.1).

The acidity or alkalinity of the environment (pH) can affect pathogen survival, with certain pathogens being more resilient under specific pH conditions. Pathogens may have different survival rates depending on the availability of oxygen in the environment. High levels of ammonia, often found in animal manures, can affect pathogen survival. Overall, understanding the factors influencing the persistence of pathogens in manures and water environments is crucial for implementing effective strategies to mitigate the risk of infection transmission to humans and animals. Proper management practices, such as composting, treatment, and disinfection, can help in reducing pathogen levels and minimize the risk of disease outbreaks (Jones, 1976; Guan and Holley, 2003) [7, 5].

Table 2: Estimation of changes in various indicators of the raw poultry and the fermented poultry waste manure

Parameters	Raw poultry manure	Fermented poultry manure
pH	8.7±0.1	4.0±0.5
Dry Matter	79.3±3.5	45.5±6.4
Ash	20.1±1.5	20.2±3.5
Crude proteins	24.6±2.6	22.9±3.4
Non protein Nitrogen	0.15±0.1	0.36±0.5
Total Volatile Nitrogen: Proteins	4.9±0.2	0
Reducing sugar	1.2±0.1	4.5±2.1

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