

# ISOLATION OF DIFFERENT BACTERIA FROM CHICKEN FEACES IN DISTRICT MANSEHRA, REGION, KPK

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## ABSTRACT:

The economy of Pakistan is generally dependent on the poultry sector after agriculture. Poultry waste litter is mainly added to the soil as a fertilizer. The last step in the poultry management policy poses a great risk to the environment due to nutrients and microorganisms contained in high concentrations in these waste materials. To find out the prevalence of microbes in poultry industry research were performed in district Mansehra of Pakistan. This study was conducted in district Mansehra region during the month of July to November 2018 to evaluate the frequency of different bacteria in the fresh chicken faeces at different poultry farms of Dhodyal, Buffa and Shinkyari region of district Mansehra. In this study total of 204 fresh faeces samples were collected from poultry farms of district Mansehra region and then isolation and identification of the pathogenic bacteria were done by culturing and by different biochemical tests. 204 fresh faeces samples of chicken were collected out of which 134 from Dhodyal, 40 from Buffa, and 30 from Shinkyari, the percentage of overall isolated bacteria in these three regions was 70.6%, 70%, and 80%. The number of bacteria was isolated and identified as *E. coli*, *Salmonella*, *Shigella*, *Klebsiella*, *Proteus*, *Pseudomonas*, *Staphylococcus* and *Streptococcus*. The frequency of isolated bacteria in these three regions was *E. coli* (31.6%, 30%, 40%), *Salmonella* (10%, 17%, 13%), *Shigella* (5%, 10%), *Klebsiella* (5%), *Proteus* (3.37%), *Pseudomonas* (5%, 5%, 10%), *Staphylococcus* (7.46%, 7.50%, 20%), *Streptococcus* (5.96%). The current study endorsed appropriate information spreading to farmers and poultry feed producers about the public health significance of proper poultry litter disposal.

**Key words:** Microorganism isolation from poultry industry of Hazara Division, Pathogenic Microorganisms.

### **Introduction:**

Poultry farms are used for raising chickens, turkeys, ducks, and other birds for egg or meat production. Poultry sector play very essential role and fulfill the requirement of protein deficiency. In Pakistan the commercial poultry sector was started in 1960. The government positive policy and the poultry farming community persistence was the result of initial growth of poultry industry (Sadiq, 2004). Although poultry are affected by enteric illnesses, this illness is one of the most important groups of diseases and is continuing to cause high economic losses in many areas around the globe due to increased death rates, decreased weight gain, increased medication costs and increased feed change rates. Several pathogens (Viruses, bacteria and parasites) are involved as possible causes of enteric disorders either alone (mono-causal), in synergy with different other microorganisms.

Before 1963 in Pakistan native chicken provided the source of egg and meat and these birds at the age of four-month yield an average 30 eggs and 0.769 kg of meat per annum (Sahota and Bhatti, 2003). These birds have been raised as backyard activities to fulfill the individual domestic desire.

The consumption of chicken meat is gradually increasing globally, the previous history and available information show that it reached to 14.2 kilogram per person, per annum (Rouger *et al.*, 2017). In 2010 the market share of mutton and beef had reduced to 20% to 55% while the poultry meat had increased to 25% respectively (Hussain *et al.*, 2015).

### **MICROBIOLOGY OF POULTRY/ CHICKEN:**

Different microorganisms such as protozoan, fungi, algae, protists, archaea, and bacteria are the dynamic part of microbial diversity and these are usually briefly mentioned or not at all which requires consideration. Microorganisms are distributed throughout the biosphere. Bacteria like to live in an energy rich environment (Ganesan and Muthuchelian, 2009). The caeca of the gastrointestinal tract of poultry is one of the favorite places for bacteria which presented favorable environment for more than 200 bacterial strains. The species of *Lactobacillus*, *Enterococcus* and *Enterobacteriaceae* are the major microflora of chick cecum at the first day of

age. After second week of the chicks age, the species of Eubacteria and bacteroides species got established (van der Wielen *et al.*, 2001). Amongst the three regions of gastrointestinal tract of chicken such as duodenum, cecum and ileum, duodenum has the lowest bacterial population while cecum has highest population of bacteria. The account of lactobacillus average is about  $1 \times 10^9$  in the ceca of chicken, along with lactobacillus huge amount of Enterococcus and Enterobacteriaceae were found. At the age of 12 day of chicks the account of facultative anaerobic and obligate anaerobic bacteria is 10 to 15 time more than aerobic bacteria. (Zhu *et al.*, 2002).

The enteric bacteria in the Enterobacteriaceae family are the major pathogen and the chicken may also be asymptomatic carrier and shed of these pathogenic bacteria in their faeces, thereby contaminating the environment. (Kilonzo-Nthenge *et al.*, 2008; Obi *et al.*, 2008) The rich nutrient contented of poultry dropping provide energy for potentially harmful organism (Chang *et al.*, 2004).

The family Enterobacteriaceae, including Klebsiella, Escherichia coli and Salmonella species are a major or secondary pathogens of poultry production. (Kilonzo-Nthenge *et al.*, 2008; Obi *et al.*, 2008).

In chicken farm administration, one of the most tough phase is the administration of the newly flock introduced. For the process to be money-making, a good disease prevention program should be offered for the newly submitted chicks to evade any future losses. Disease can be spread by human, other birds, newly introduced chicks or contaminated gear. Disease controlling from the beginning is the significant for the success of the operation (Mobley and Kahan).

## **STUDY AREA**

District Mansehra has a large number of poultry farms and plays very important role in meat production. The peoples of this area mostly depend on poultry. On the other hand, the effect disease in chickens before of this has never been studied in that area. The environmental and management circumstances are the main causative points which can greatly increase the incidence of poultry in District Mansehra and probable area would be focused. Correspondingly, in recent past, antimicrobial susceptibility test was mostly suggested for the investigation and

observing of antimicrobial resistance in different areas continuously. Hence, this study is designed to find the prevalence of different bacteria in chicken faeces in this area and to found the antimicrobial sensitivity profile against the pathogens related with poultry.

### **AIMS AND OBJECTIVES**

To isolation and identification of bacterial consortium in chicken faeces samples from collected in Mansehra Region.

### **MATHODOLOGY:**

#### **SAMPLE COLLECTION:**

A total 204 fresh chicken faeces samples were collected from different poultry farms of District Mansehra region. The samples of fresh chicken fecal matter were taken randomly from chickens of different health status by using sterilized cotton swabs and transported to the laboratory.

#### **Transport Media**

Different transport media are used for delivering fecal samples to laboratories such as Campy-thioglycolate, Carry Blaie, Semi-solid motility test medium and Alkaline peptone water. In the current research, sterilized cotton swab was used for transporting chicken fecal samples (without contamination and disruption of samples taken with culture swabs) to the laboratory Of microbiology Department Hazara University within two hours for isolation and identification of different bacteria.

#### **TO STERALISE PETRI PLATES AND AS WELL AS TEST TUBES:**

In drying method, we sterilized the petri plates as well as the test tubes with tap water and also kept in hot air oven. Then autoclaved the petri plates and the test tubes for 15 minutes at 121 °c under 15 psi pressure. For further process the petri plates and the test tubes were kept in Luminar flow hood.

#### **Culture media preparation:**

The preparation of media was done according to the labeled requirement given on the bottle. For the ingredient of each media separate flask were used and then mix the ingredient by heating and

shaking. Then the flasks were covered by aluminum foil and autoclaved. After autoclave the media flask were kept to cooled. In the blood agar flask blood were added and gently shake to mix the blood in agar (hours and sheep blood). Then the Luminar flow hood was washed with ethanol and on the ultra violet light for 10 minutes before the media was transferred.

### **INCUBATION PERIOD FOR PETRI PLATES:**

The incubation period up to twenty-four hours (24 hos) for petri plates given to eliminate the microbe's contamination because for further process no microbial contamination occurs on the petri plates.

### **Sample processing:**

Sterilize cotton swab which is taken from chicken faeces were directly inoculated on blood agar, MeConkey agar, and Salmonella, Shigella agar (SS agar), for the detection of different bacterial colony. The petri plates were kept in in reversed position at 37°C for one day and after the incubation period the colony were appeared on some plates.

### **Purification:**

The colony on MeConkey agar, S.S agar and blood agar were obtained and identified on the basis of morphology and biochemical properties. For obtaining of pure colonies again I cultured of already isolated colonies and kept in incubator at 37°C for 24 hours.

### **Morphological identification:**

### **Colonial Identification:**

After 24 hours' incubation bacterial growth were occur in colonial form. On the basis of colony, surface texture, color, size, elevations, shape and edges different bacterial colonies were identified. Two types of colonies were formed on Mac-Conk agar (a) Grayish (b) yellowish. Those colonies which are yellowish were identified as Lactose fermenter and those colonies which are grayish in color identified as the Non-lactose fermenter. On blood agar white grayish colony of different bacteria were appeared which have different shape, size and some bacteria such is streptococcus species showed hemolysis on blood agar which may be alpha, beta and gamma hemolysis, while two different types of colonies were appeared on SS agar. Opaque

colorless colonies with black center were identified as Salmonella and Smooth round transparent colorless with no black center colonies were identified as Shigella. Further morphological identification of different bacterial colonies was done through gram-staining.

### **GRAMSTAINING AND MICROSCOPY:**

For gram staining process, a clean slide was taken and then up to one drop of normal saline was dropped on the slide. A well sterilized wire loop was used to make a smear on the slide and the slide was dried and fixed through heat. The crystal violet was poured on the slide and the incubation period for 1 minute and 30 seconds and washed the slide properly with tap water. After it we used the gram iodine for 1 minute and 30 seconds and then washed again with tap water. After the gram iodine we used alcohol up to 95 % for 15 to 20 seconds and then washed with tap water. Then we used the safranin for 1 minute and then again washed with water as well. Now we used to dry the slides in normal air and then we examined the slide under a microscope. A thick layer of the peptidoglycan-90% present in the cell wall of gram positive bacteria and the cell wall-stain with purple color and while a thin layer of peptidoglycan-10% present in the cell wall of gram negative bacteria and the cell wall were stained with red or pink color. Further, identification of bacteria was done on the basis of biochemical test such as catalase, oxidase and amvis test.

### **Biochemical identification**

#### **METHOD FOR CATALASE TEST:**

There are two methods are used for catalase test which are given below

#### **A: TUBE METHOD:**

- 1:** Poured 1-2 ml of Hydrogen peroxide solution mix in a test tube.
- 2:** pick of several colonies from the 20 to 24 hours' incubation period of culture medium with the help of sterile wire loop and dip in the solution of Hydrogen peroxide.
- 3:** Examine the abrupt producing of oxygen bubble occurs inside a test tube.

#### **B: SLIDE METHOD:**

- 1:** Pick the small colonies from the growth by using the sterile wire loop and placed the colonies on a well cleaned and dry glass slide.

2: Mix 3% of Hydrogen peroxide with colonies on the slide.

3: Examine the bubbles of oxygen produced.

### **PROCEDURES OF OXIDASE TEST:**

This test was used for those bacteria which have an enzyme cytochrome oxidase.

1: This test was also used for the differentiation of bacteria like *Campylobacter*, *Moraxella*, *Neisseria*, *Pasturella* and as well as *Moraxella*.

2: It is mainly used to differentiate pseudomonas species from the co related species.

### **IMVIC TEST:**

#### **REAGENT FOR IMVIC TEST:**

##### **Methyl Red:**

Distilled water 500ml, Ethyl alcohol.

##### **Method for preparation:**

in 300 ml ethyl alcohol, Methyl red was dissolved then distilled water was added. For each test Five drops of above reagent were used (Distinct red color is positive test. Yellow color is negative reaction).

##### **Vogues -Proskour Reagents:**

KOH (40%) (0.2 ml) Naphthol solution (0.6 Alpha – Naphthol)

**Method of preparation:** (VP) broth tubes were Inoculated by suspected colonies, incubated for  $48 \pm 2$  hr. at  $35\text{ C}^\circ$ , then 0.6 ml L- Naphthol solution and 0.2 ml 40% KOH (potassium hydroxide) solutions were added then mixed well. Eosin pink color is considered positive.

**Kovacs reagent, (ICMSF,1978):** Para dimethyl amino benzaldehyde 5.0 g, Isoamyl or (normal amyl) alcohol 75.0 ml Hydrochloric acid (cone.) 25.0 ml.

**Method of preparation:** Benzaldehyde was dissolved in isoamyl alcohol then HCL conc. was added slowly, 0.5 ml of above mixture was used for each indole test.

### **BIOCHEMICAL TESTS:**

### **Methyl Red test:**

Five ml of culture in MR-VP both was incubated at 37C° for 24-48h. Five drops of methyl red solution were added to each tube. Distinct red color is a positive test while a yellow color shown as a negative test.

### **Vogues- Proskour test:**

MR-VP broth was incubated at 37C° for 24-48h and then a Few drops of reagent alpha-Naphthol solution and 0.2ml 40% KOH were added and mixed, pink color development was considered positive.

### **Indole test:**

Five ml of culture in peptone water was incubated at 37C° for 24-48h. Indole production was tested by adding 0.5 ml of Kovacs, reagent. Appearance of distinct rosy colour in upper layer is appositve test.

## **RESULTS**

In the present study, was conducted to investigate different pathogenic bacteria from chicken faeces in three different regions of Mansehra district. Total samples of chicken faeces were 204, that were collected from different poultry form. Out of these, 134 sample were collected from dhodyal region,40 sample from Buffa region and 30 samples were collected from shinkyari region. All the samples were conformed primarily with the help of its growth characteristics on three different media such as Blood agar, Meconkey agar and Salmonella Shigella agar and then isolate were identified with the help of biochemical tests.

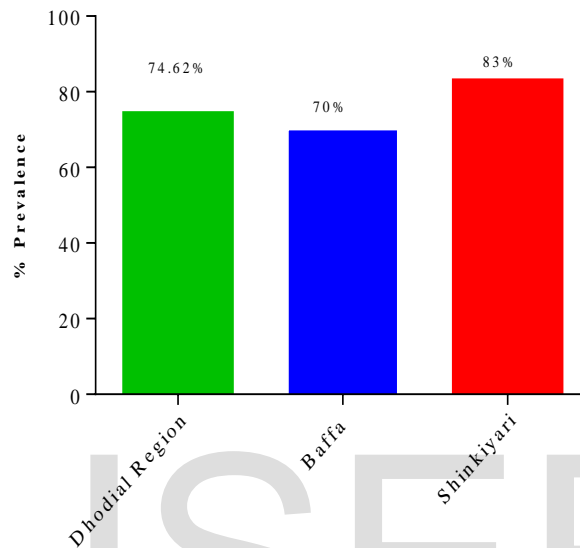
### **Prevalence of pathogenic bacteria in three different regions of District Mansehra:**

204 fresh faeces sample were collected from three different regions of Mansehra which include Dhodyal, Buffa, and Shinkyari for the detection of different pathogenic bacteria.134 samples were collected from Dhodyal region, 30 samples from Shinkyari and 40 samples were collected from Buffa region. High incident of bacteria were shown in the samples which were collected from Shinkyari region. 25 samples out 30 were positive and shown high percentage of 83% followed by Dhodyal where 102 out of 134 samples were positive in result showed 74.62%. In



Buffa region its percentage was quite low where 28 out of 40 samples were positive and having a percentage of 70%. From three different regions the percentage of positive bacteria were isolated as shown (fig 3.1)

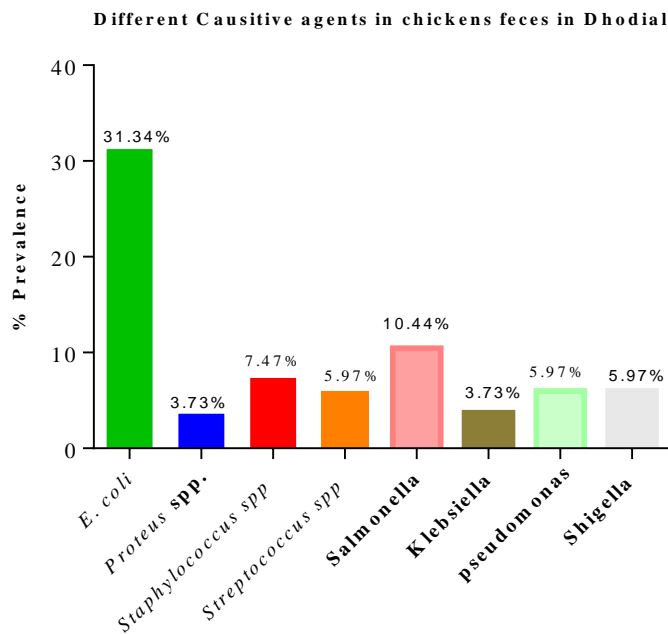
ISOLATION AND IDENTIFICATION OF PATHOGENIC BACTERIA FROM CHICKEN FACES IN DISTRICT MANSEHRA REGION KPK.



**Fig.3.1: Percentage distribution of various pathogenic bacteria screened out in chicken feces from Dhodial, Buffa and Shinkyari region.**

**Prevalence of different causative agent in Dhodial region:**

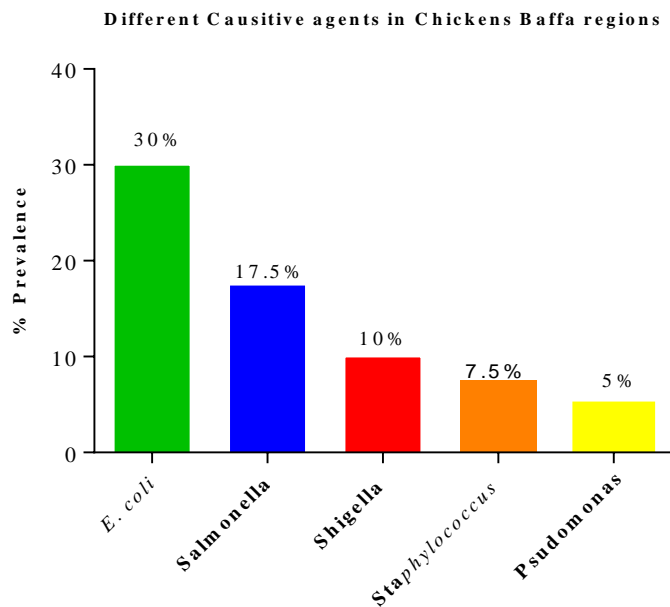
Fresh faeces sample of 134 were collected from different poultry farm of Dhodial region, 10 to 13 samples were collected from each farm for the detection of different bacteria. Out of 134 samples, 102 were positive and the remaining 32 samples were found to be negative. The incidence rate of different bacteria was shown in the (fig 3.2).



**Fig 3.2: Percentage distribution of various pathogenic bacteria screened out in chicken feces from Dhodyal.**

### **Prevalence of different causative agent in Buffa region:**

40 fresh faeces samples were collected from four poultry farm of Buffa region, 10 samples were taken from each farm for the isolation of various bacterial pathogens. Out of 40 samples, 28 samples were found positive after culture on three different medias. and the reaming 12 samples were not shown any bacterial growth on medium plates. Hence the incidence rate of different bacteria was shown in the (fig 3.3).

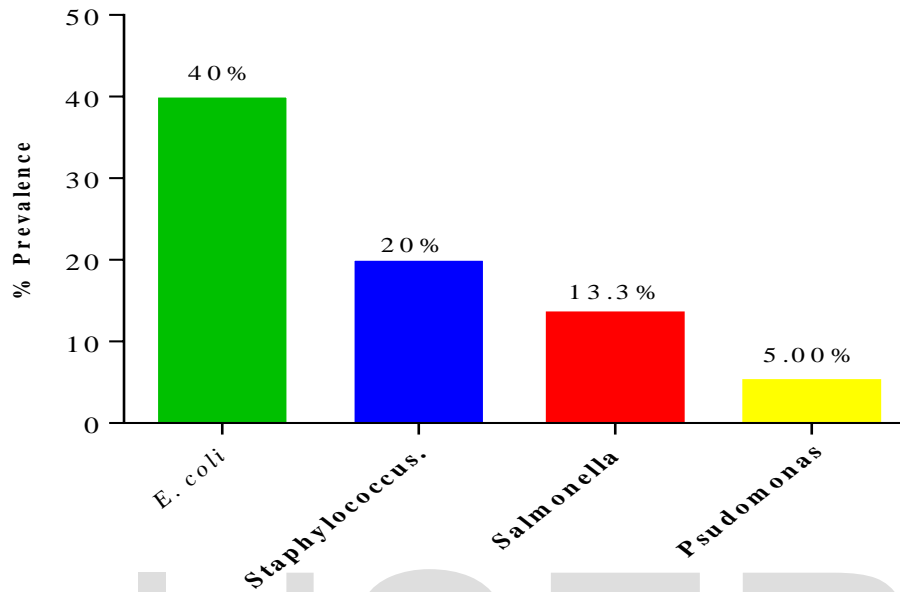


**Fig 3.3 Percentage distribution of various pathogenic bacteria screened out in chicken feces from Buffa.**

**Frequency of different causative agent in Shinkyari region:**

From three different poultry farms of Shinkyari region, 30 fresh faeces samples were collected, 10 samples were taken from each farm for the identification of different bacterial species. Out of 30 samples, 25 samples were shown positive results after culturing on different medias. And the remaining 5 samples were found to be negative. The incidence rate of different bacteria was shown in (fig 3.4).

Different causitive agents in chickens Shinkiyari region

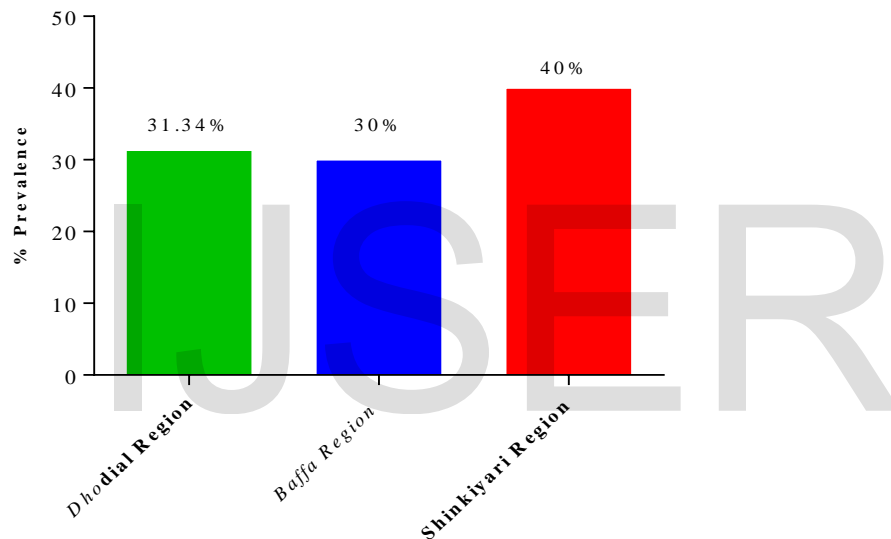


**Fig 3.4: Percentage distribution of various pathogenic bacteria screened out in chicken feces from Shinkyari region.**

### Frequency of *E. Coli* among chicken faeces in different regions of district Mansehra:

204 fresh faeces sample were collected from different poultry farms of Dhodyal, Shinkyari and Baffa regions of district Mansehra. In shinkyari, the incidence rate of *E. coli* was found to be 40% which was higher, followed by Dhodyal 31.34% and Baffa region it was found 30% as shown in the (fig 3.5).

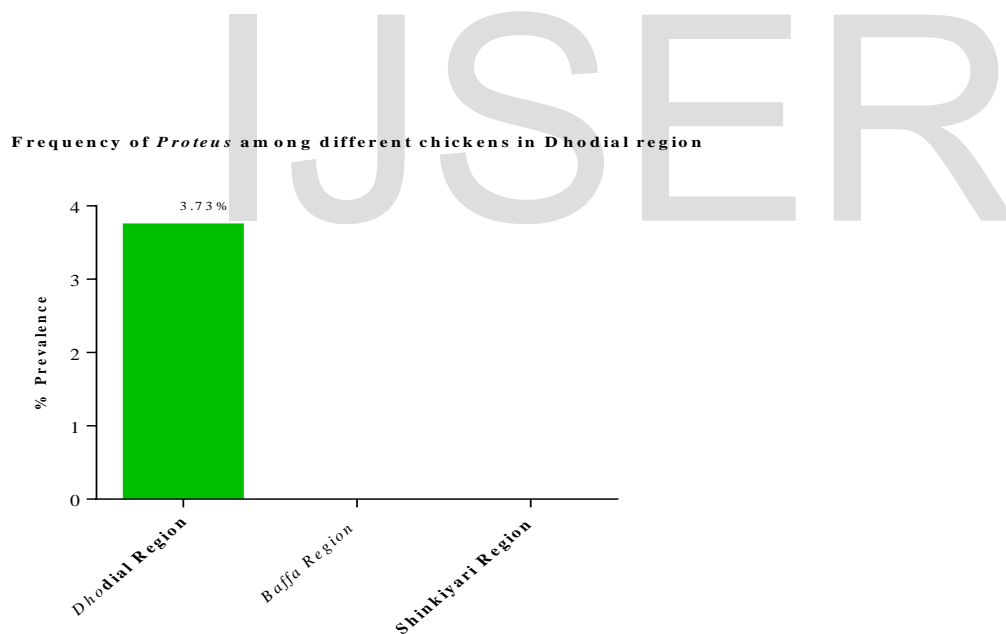
Frequency of *E. coli* among chicken faeces in different region of District Mansehra.



**Fig 3.5: Percentage distribution of *E. coli* among chicken's feces in Shinkyari, Dhodyal and Baffa regions.**

### Frequency of *Proteus* among chicken in different region of Mansehra district:

Faecal samples of chicken were collected from Dhodyal, shinkyari and buffa region of district Mansehra. Out of 204 samples, *Proteus* was identified only in Dhodyal region. The ratio of *Proteus* in Dhodyal region was 3.73% as shown in the (fig 3.6).

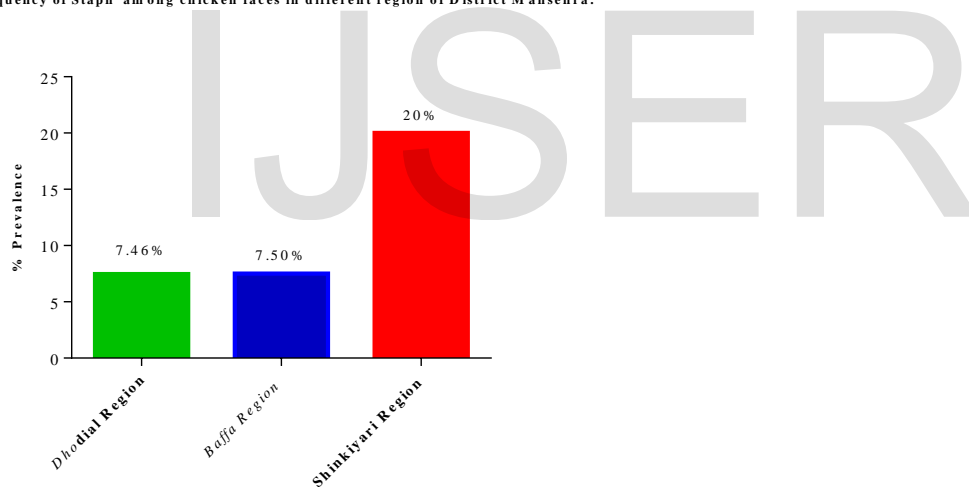


**Fig 3.6: Percentage distribution of *Proteus* among chicken's feces in Dhodyal**

### Frequency of staphylococcus species among chicken faeces in different region of Mansehra district:

204 faecal samples were collected from Dhodyal, Shinkyari and Buffa region of district Mansehra. The ratio of Staphylococcus species in Shinkyari was higher which percentage was (20%), followed by Buffa which percentage was (7.50%) and Dhodyal (7.46%) as shown in the Fig 3.7.

Frequency of Staph among chicken faces in different region of District Mansehra.



**Fig 3.7: Percentage distribution of *Staphylococcus* species among chicken's feces in 3 regions of district Mansehra.**

### Frequency of *Streptococcus* among chicken feces in different region of Mansehra district:

Faecal samples of chicken were collected from Dhodyal, shinkyari and buffa region of Mansehra district. Out of 204 samples *Streptococcus* was identified only in Dhodyal region. The ratio of *Streptococcus* in Dhodyal region was 5.97% as shown in the (fig 3.8).

Frequency of *Streptococcus* among chicken faces in Dhodial region



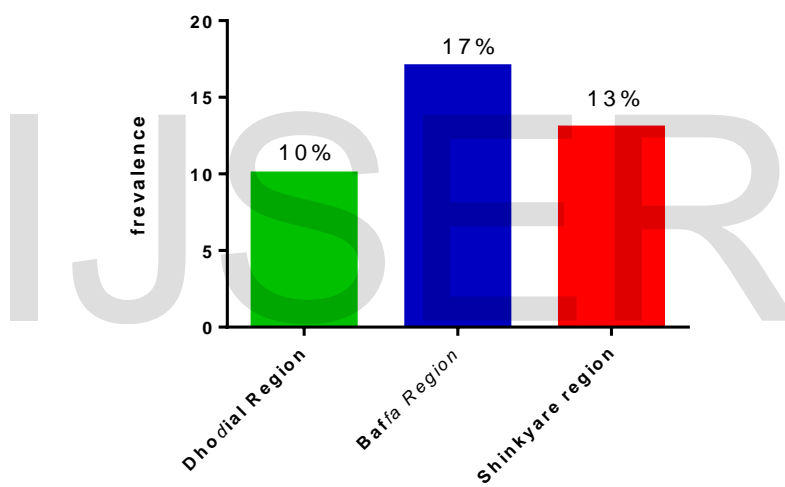
**Fig 3.8: Percentage distribution of *Streptococcus* among chicken's feces in Dhodyal.**



### Frequency of *Salmonella* among chicken feces in District Mansehra:

204 faecal samples of chicken were collected from different regions that was included by Dhodyal, shinkyari and buffa region of district Mansehra. The ratio of *Salmonella* in Baffa region was higher as (17%) compared to other two regions. Shinkyari was recorded as (13%) and Dhodyal was (10%) as shown in the Fig 3.9.

Frequency of *Salmonella* among chicken faces in District Mansehra region

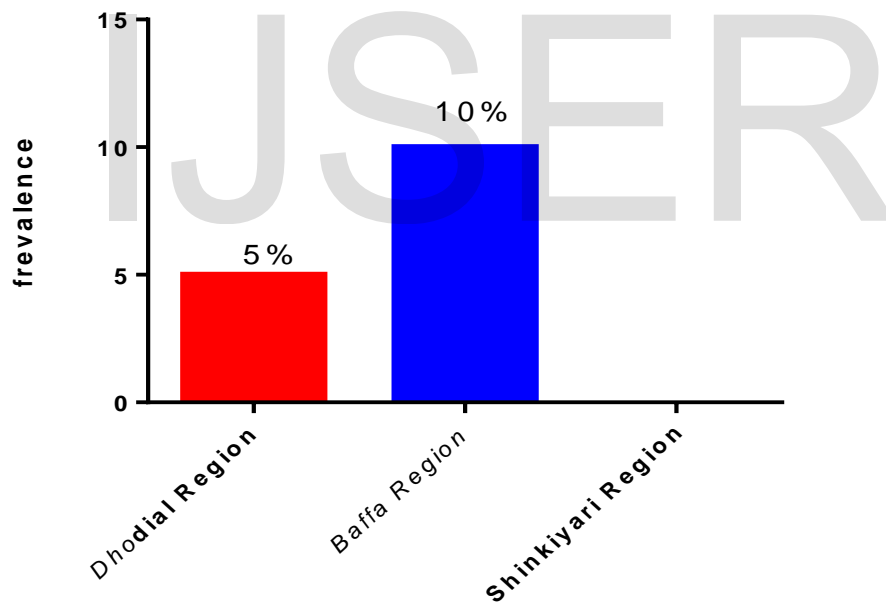


**Fig 3.9. percentage distribution of *Salmonella* among chicken's feces in in district Mansehra region.**

### Frequency of *Shigella* among chicken feces in District Mansehra region

From Dhodyal, shinkyari and buffa region of district Mansehra, 204 chicken fresh faecal samples were taken. The ratio of Salmonella in Baffa was higher as (10%). Followed by Dhodyal which was (5%). *Shigella* was not identified from the region of Shinkyari as shown in the Fig 3.10

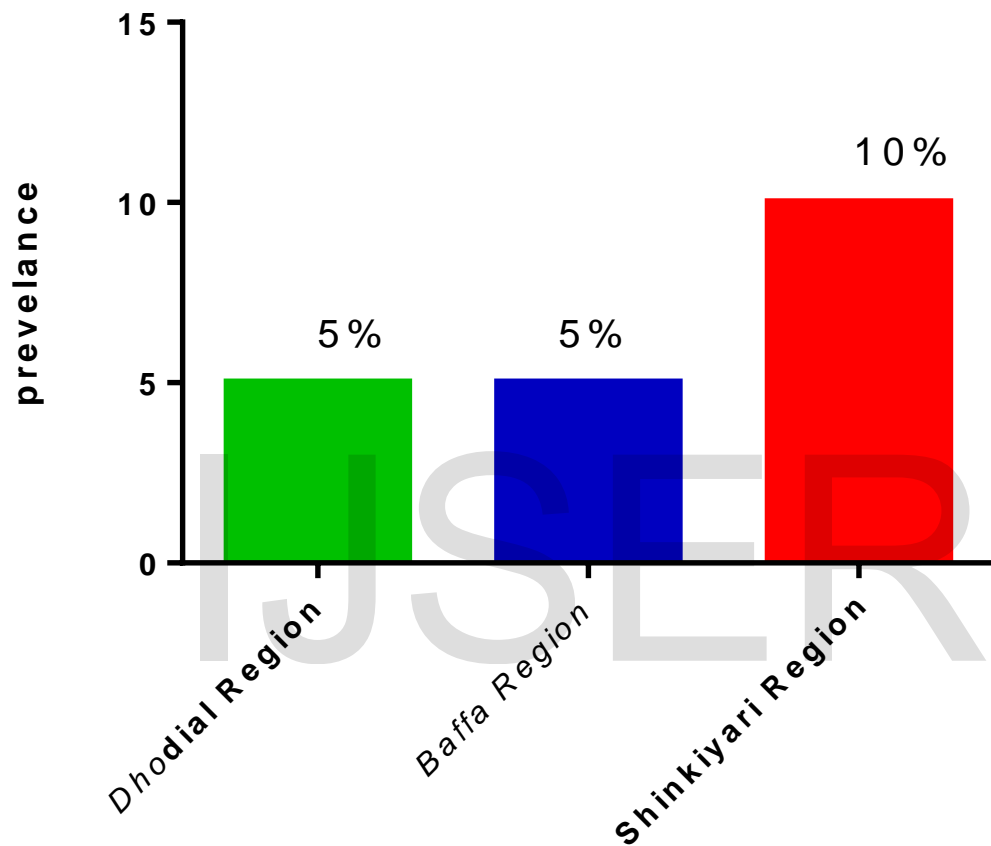
Frequency of *Shigella* among chicken faces in District M ansehra region



**Fig 3.10: Percentage distribution of *Shigella* among chicken's faeces in Dhodyal, Baffa regions.**

### Frequency of *Pseudomonas* among chicken feces in District Mansehra region:

204 faecal sample of chicken were taken from different poultry farm of Dhodyal, shinkyari and buffa region of district Mansehra. The ratio of *Pseudomonas* in Shinkyari was higher as 10% than other two regions. In Baffa region it was found as (5%) and same percentage was found in Dhodyal as shown in (fig 3.11)

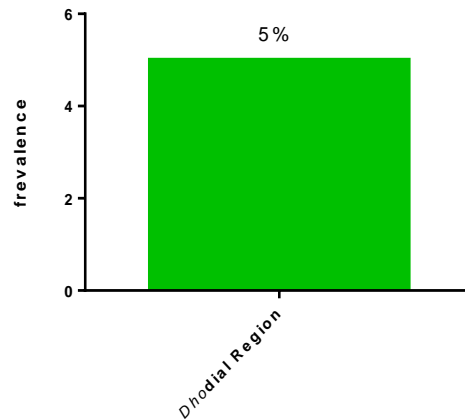


**Fig 3.11: Percentage distribution of *pseudomonas* among chicken's faeces in Dhodyal, Baffa, and Shinkyari region.**

#### **Frequency of *Klebsiella* among chicken feces in different region of Mansehra district**

From different poultry farm of Dhodyal, shinkyari and buffa region of district Mansehra, total 204 fresh chicken faecal samples were collected. Out of these samples *Klebsiella* were identified

only in Dhodyal region. The ratio of *Klebsiella* was 5 % in Dhodyal region as shown in the (fig 3.12).



**Fig 3.12: Percentage distribution of *Klebsiella* among chicken's faeces in Dhodyal, region.**

## DISCUSSION

In Pakistan poultry is an important agricultural industry and having an investment of more than 732 billion rupees. Poultry meat contained enough amount of protein having various important amino acid like methionine, histidine, leucine, cysteine, arginine, and isoleucine etc. (Uddin *et al.*, 2018). Poultry meat is appreciated product for the consumer and their faeces are used as fertilizer in District Mansehra region. The farmer used the chicken faeces is good source of manure for the cultivation crops and vegetable. The use of poultry dropping for the cultivation of crop serve the dual purpose elevating the soil for enhanced crops yield and economically disposing of the dropping. However, the directly addition of poultry dropping to field without any form of treatment possess some public health problem since they contain pathogenic microorganism. The pathogenic microorganism can contaminate the surrounding crops and vegetable and become the source of infection especially when such crops or vegetable are eaten raw or brought home where they can contaminate other materials. The pathogenic

microorganism can also be discharge onto surface that are used for drinking after run-off during rain fall.(Orji *et al.*, 2005).

The present study was conducted to investigate the prevalence of pathogenic bacteria from chicken faeces in three different regions (Shinkyari, Dhodyal, and Baffa) of district Mansehra. However, there are limited report were available from Pakistan. However, no such work has ever been done in Hazara division, especially in District Mansehra region. Out of total 204 samples analyzed, 155 samples were positive showing a high prevalence of pathogenic bacteria in poultry i.e. 75.98%.

Among positive cases in three different regions, highest incidence of bacteria was shown in Shinkyari region and least incidence in buffa region with percentage of 83% and 70% respectively. Whereas, the prevalence of bacteria in Dhodyal region was appeared 74.62%.

Pathogenic bacteria isolated in the present study included *E. coli*, *Salmonella*, *Shigella*, *Proteus*, *Staphylococci*, *Pseudomonas*, *Klebsiella*, and *Streptococci*. The *E. coli*, *Staphylococci*, *Salmonella* and *Pseudomonas* species were isolated from all three regions with highest prevalence in Shinkyari region compared to Dhodyal and Baffa. Contrarily, *Proteus*, *Streptococci* and *Klebsiella* species were only found in Dhodyal region and *Shigella* in Dhodyal and Baffa region of district Mansehra.

*E. coli* was found most prevalent among all the isolated bacteria in selected regions probably because of its commonness in the normal microflora of intestine. Its prevalence in Dhodyal, Shinkyari and Baffa regions was 31.34%, 40% and 30%. In contrast to present study, 55.50% *E. coli* was isolated from chicken dropping in Nigeria in 2017 (Ajayi and Omoya, 2017).and Similar study was done in Kenya and isolated *E. coli* from poultry dropping with the prevalence rate of 57% (Langata *et al.*, 2019). while in 2015 42% *E.coli* was isolated from cloacal and liver of broiler chicken(Khatun *et al.*, 2015).In 2016 *E.coli* isolated from commercial broiler and backyard poultry in Lahore Pakistan with prevalence rate of 64.2 and 54.5.(Akhtar *et al.*, 2016)

The *Salmonella* species, after *E. coli*, were found more prevalent in Shinkyari (13.3%) and Baffa (17.5%) regions compared to Dhodyal where its prevalence was only 10.44%. A study was steered in Faisal Abad Pakistan and isolated *Salmonella* from poultry dropping with percentage

of 55% (Akhtar *et al.*, 2010) while 5% was shown by (Mohammed and Ibrahim, 2012).and 21.4% 32% was shown by the report,(Bhuvanewari *et al.*, 2015).

Similarly, the prevalence of *Staphylococci* was highest in Shinkyari region i.e. 20%, followed by Baffa and Dhodyal region where its prevalence was 7.50% and 7.46%. The percentage *S. aureus* isolated from this study was less than 24.4% Occurance reported by (Bala *et al.*, 2016) and more than (Pesewu *et al.*, 2018) isolated coagulase negative *S. aureus* from chicken meat with the percentage of 9.2%.

The *Shigella* was only isolated from Baffa and Dhodyal regions of district Mansehra. The ratio of *Shigella* in Baffa region was 10% followed by 5% in Dhodyal region. Whereas, it was not identified in chicken fecal samples from Shinkyari region. the previous study reported in Saudi Arabia Taif and isolated *Shigella* from Cloacal area of poultry with the percentage of 18%.(Abo-Amer and Shobrak, 2015) Which were higher from my study.

The *Proteus*, *Streptococci* and *Klebsiella* species were only found in Dhodyal region. Their prevalence was reported as 3.73%, 5.97%, and 5% respectively. In addition to their isolation only from Dhodyal region, the *Proteus* and *Klebsiella* were the least prevalent species among all the isolated bacteria from this region. Contrast to my study (Jambalang *et al.*, 2017) isolated *Klebsiella* and proteus from the chicken egg in South Africa, and (Nahar *et al.*, 2014) isolated proteus from chicken dropping with the percentage of 39%. While (Siddiqui *et al.*, 2008), isolated *Streptococcus* from heart blood, lung, gallbladder intestine and faeces of birds with percentage of 6.796%. the difference from my study is due to diagnosis, different environment and geographical variation.

*Pseudomonas* species were isolated from all three regions of district Mansehra. Their prevalence in Shinkyari region was highest (10%), followed by Baffa and Dhodyal region where *Pseudomonas* was equally prevalent i.e. 5%. Moreover, the prevalence of *Pseudomonas* in Baffa and Dhodyal region was lowest among all the isolated bacteria from these two regions. In contrast to my research 5% *pseudomonas* species isolated from meat in Iraq (Noori and Alwan, 2016)

## CONCLUSION:

The current result provided indication that poultry waste can serve as an environmental reservoir for multiple antibiotic resistance bacteria and hence can serve as possible routes for the entrance of multidrug resistance zoonotic pathogen in the human being. This has very significance inference for human health, as multidrug resistance infections were problematic to treated and frequently required costly antibiotic and long term therapy. This can considerably rise the cost of treatment and even death. The study consequently endorsed appropriate information dissemination to agriculturalist and poultry feed maker about the public health prominence of proper poultry waste disposal.

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