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ISOLATION, IDENTIFICATION AND ANTIBIOGRAM OF *SHIGELLA* IN BROILER POPULATION RAISED IN DISTRICT ABBOTTABAD

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Abstract

Shigella is a Gram-negative, non-motile, rod-shaped bacterium belongs to family Enterobacteriaceae. Shiga toxin is a toxin produced by Shigella species strains. Shigellosis is regarded as a major issue for both public and veterinary health. Shigella is the cause of bloody purulent diarrhea and food poisoning in humans. It also causes several animal illnesses and has a high death and morbidity rate. Shigella spp. are disseminated by direct contact with an infected person, contaminated food, or polluted water. A cross-sectional survey was conducted to determine the occurrence of Shigella in broilers from different poultry farms from different locations in District Abbottabad and to determine their antibiogram. In the present study, overall 200 liver samples of dead chicken during October 2017 to April 2018 were used to check the isolation identification and antibiogram of shigella. Shigella was isolated after passing the samples through pre-enrichment, selective enrichment and culture in a selective medium and identified using standard microbiological and biochemical methods. Antibiotic susceptibility test was performed by the disc diffusion method according to the CLSI method (Kirby-Bauer disk diffusion test) on Muller-Hinton agar. Liver samples were collected from broiler chicken from five age groups and from seven different areas of district Abbott bad. All these samples were examined by traditional method. After examination 36/200 sample were positive for shigella with incidence of 18%. Samples were collected from different age-groups. After isolation, a high incidence was found in 8-14 days old broiler (25%) rather than 29 -35 days and 0-7 days (19%, 16%) respectively, and a low incidence 22-28 days 15-21 days and (11%.12%) was found. Total 36 isolates were tested for antimicrobial drug susceptibility against 5 commonly used antibiotics belonging to different groups by disc diffusion method or Kirby-Bauer method. Antibiotic sensitivity and resistance patterns of isolated Shigella spp. were studied by disc diffusion method using 5 commonly prescribed antibiotics. Shigella showed resistance to various antimicrobial agents at variable degree. It was observed that the most sensitive antibiotic is Gentamicin. Ciprofloxacin and Amoxicillin were found to be most resistant drugs and Enrofloxacin and Colistin sulfate are intermediate. These findings would certainly help the veterinarians to select the correct antibiotics against Shigella infections.

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INTRODUCTION

Poultry may be defined as birds that produce both meat and eggs. In terms of animal production operations, the poultry industry is expanding quickly. It can provide disadvantaged farmers with resources for revenue and the opportunity to feed the fastest-growing population (Ogunlade *et al.*, 2017). In Pakistan, the poultry industry contributes significantly to both the supply and demand for protein. Commercial poultry operations in Pakistan began in the early 1960s and showed signs of rapid expansion over time. This was the outcome of the poultry farming community's perseverance and the early development of this sector of the management's commendable and advertising campaigns. On a commercial level in Pakistan When Pakistan International Airlines (PIA) established the country's first contemporary hatchery in Karachi in 1963, poultry farming emerged (Ullah *et al.*, 2019). The poultry business is the nation's second largest industry, producing 530.00 million birds annually (Hussain *et al.*, 2015).

Pakistan's chicken industry has historically and now encountered several illnesses. Diseases caused by bacteria, viruses, and parasites have a negative impact on poultry productivity. Bacterial infections are the cause of delayed development and a poor feed conversion ratio (Aslam *et al.*, 2020). The Gram-negative bacterium Shigella, sometimes known as shigellosis, is an infectious and management issue in the production of broiler chickens. when Shigella and EIEC infections are present. Shigella is a facultative anaerobe, meaning that it may grow anywhere from 6°C to 48°C. Shigella can survive for 5–10 days in acidic foods like orange and lemon juices and for 50 days in neutral foods like milk, wheat, and eggs (Lampel *et al.*, 2018). Their pH range is 4.8–9.3. *Escherichia coli* and Shigella are closely related, and Shigella is a representative member of the Enterobacteriaceae family. Strain D, *S. sonnei*, is the primary cause of Shigella-related diarrhea (shigellosis), and all four of these species have distinct and varying levels of pathogenicity. Kiyoshi Shiga made the discovery of this genus in 1897. It is a novel agent of human shigellosis and is only found naturally in humans (Lampel *et al.*, 2018).

Low feed quality, cold stress, inadequate ventilation, and humid conditions that encourage coccidiosis are the main causes of shigellosis. According to the CDC's evolving Infections Program, Shigella species were the third most common food-borne bacterial pathogen reported in 2002 (Shi *et al.*, 2014). A particular kind of gastrointestinal bacterium called Shigella causes illness in both humans and other primates. Shigellosis, an infection brought on by ingesting the Shigella bacterium, is typically accompanied by diarrhea and other stomach symptoms. It is commonly known that food plays a part in Shigella transmission. After 30 days, Shigella may be recovered from milk, cheese, eggs, and shrimp at room temperature. Similarly, cheese is a typical source of contaminated dairy and milk products (Sackey *et al.*, 2001).

According to epidemiological data, the most common and significant contributing factor to foodborne shigellosis cases in both industrialized and developing nations is inadequate personal hygiene. Dysentery is typically the result of infection (Warren *et al.*, 2006). Primates and other humans are often Shigella natural pretenders. Nevertheless, instances of Shigella infections in novel hosts, such as rabbits, piglets, calves, and primates, have surfaced. In order to promote infection, Shigella species should thrive in the stomach acid environment and target colon epithelial cells. The infected cells die as a result of Shigella spp. multiplying inside the colon's epithelial cells and spreading to neighboring cells. Bloody mucosal diarrhea, which is frequently indicative of a Shigella infection, results from the colon being ulcerated, inflamed, and shedding mucoid dead cells (Warren, 2006).

Antibiotics including ampicillin, ciprofloxacin, nalidixic acid, and sulfamethoxazole are used to treat shigellosis. Although Shigella can be eradicated with consistent treatment, Shigella spp. have been created by the frequent use of antibiotics and horizontal gene transfer (Das *et al.*, 2013). Shigella can readily contribute to these expansive and included actions and be redistributed and improved by a variety of paths when antimicrobial agents are routinely used on poultry for various purposes, such as

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development expansion or therapeutics. This might lead to the need for

anti-Shigella strains that are multi-drug resistance (MDR) (Ranjbar and Farahani, 2019).

MATERIALS AND METHODS

Study area and Source of specimens

The specimens were collected from sick broilers and layers that were chosen from several poultry farms and the Veterinary Research and Disease Investigation Center in Abbottabad from October 2017 until June 2018.Various farms in Abbottabad that engage in commercial broiler rearing will provide the morbid materials of broiler chickens.

Collection of samples

To isolate, identify, and evaluate Shigella susceptibility to antibiotics, a total of 100 liver samples were collected from randomly selected deceased broiler chickens from broiler poultry farms in District Abbottabad, KPK, Pakistan. Sterile plastic bags containing liver samples were promptly taken to the lab under the correct transportation conditions.

Poultry necropsy

After the afflicted birds died, the hen was placed on its back. After the legs were securely grasped in the vicinity of the femur and bent until the femur head was fractured, the two legs were placed flat on the table. After cutting the skin using surgical scissors until the whole ventral portion of the body, including the neck, was visible, the abdominal cavity was revealed by cutting the breast muscles and abdominal wall (Sharma *et al.*, 2017).

Figure 1: Poultry necropsy

Tissue samples

Tissue samples (livers) were detached with sterile scissors and forceps and retained into sterilized test tubes and bags.

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Figure 2: Tissue samples (livers)

Culture of samples

The assembled samples were inoculated and cultured on diverse culture media in one to two hours. Media used for culture are HEA agar and SS agar. HEA agar and SS agar plates inoculated were incubated at 37°C for 24-48 hours. Next given time the plates were observed, and colonies were further processed.

Culture media for identification of *Shigella*

Buffered Peptone Water

In one liter of distilled water, fifty grams of powdered agar were dissolved. After thoroughly mixing the powder, it was divided among test tubes and autoclaved for 15 minutes at 121°C to sanitize the medium. Keep it at room temperature to chill. After that, until it is utilized, keep the medium in the refrigerator at 4 °C. After carefully mixing the liver's contents, 25 g of the combined liver pieces were added to 225 ml of peptone broth. The surface-sterilized liver will be cut into pieces using a sterile blade. Careful homogeneity of the mixture was carried out. At 37°C, the pre-enriched liver samples were incubated for the whole night (Uyttendaele *et al.*, 2001).

Salmonella Shigella (SS) Agar

For the isolation and culture of Shigella species, Salmonella Shigella (SS) Agar is a differential medium with a modest level of selectivity. SS Deoxycholate Citrate Agar is transformed to become Agar. Agar powder was mixed with 1000 milliliters of distilled water in a flask. To ensure that all of the agar components had dissolved, the combined agar was heated. After that, the agar was placed in sterile Petri plates and kept there at a sterile surface until it crystallized for usage. Before inoculating, the plates were left at room temperature to warm and dry the agar surface. The pre-enriched isolate was streaked onto agar for culture using a stirring loop. For 18 to 24 hours, plates were incubated aerobically at 35 to 37°C (Nataro *et al.*, 2011).

Hekton enteric agar (HEA)

One of the primary agars for Shigella spp. growth is HE agar, which is made possible by adding a lot of peptone and carbohydrates, which reduces the inhibition of these organisms by bile salts. In one liter of distilled water, 1.75.1g of agar powder was mixed with boil for two minutes to completely dissolve the powder, stirring constantly and stirring constantly until the powder is completely dissolved. The agar surface was allowed to warm and dry on the plates at room temperature prior to inoculation. In order to cultivate the specimen, it was streaked on agar using a stirring loop. For eighteen to twenty-four hours, plates were incubated aerobically at 35 to 37°C (Nataro *et al.*, 2011).

Culture characteristics

Gram staining

Bacterial morphology is determined by Gram staining. Gram-positive bacteria are 90% coated in cellulose and a thick covering of peptidoglycan. Gram-positive bacteria exhibit crystal violet or purple coloration following the gram staining procedure, and acid alcohol did not decolorize them. Gram-negative bacteria have lipids and a thin coating of peptidoglycan covering 10% of their cell walls. After staining, Gram-negative bacteria had a red or pink color. Acid alcohol decolorizes these bacteria, which are then stained with crystal violet. When counter-stained with neutral red or safranin, the stain becomes red or pink. A sterile loop is used to select the colony, and a smear is created on a glass slide, let to dry in the air, and then fixed

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with heat. Crystal violet was used as a basic stain for 30 to 60 seconds on glass slides that had smears on them. Water may be used to clean the slide. On the slide, the iodine solution was applied for 30 to 60 seconds. With the aid of water, the slide was removed. Gram-positive bacteria maintained their blue hue whereas gram-negative bacteria were completely decolorized after a few seconds of acid alcohol treatment. The slide was once more removed with the aid of water. For 120 seconds, either safranin or neutral red stain was used. On the other hand, the slide was cleaned using water and tape. The slide was examined under a microscope using an oil immersion at a 100x magnification after it had air dried (Abdallah *et al.*, 2016).

Identification of *Shigella*

Biochemical test

Triple Sugar Iron Agar Test

The bacteria are differentiated by the utilization of sugar (fermentation) and hydrogen sulfide (production). The medium's yellow hue indicates the fermentation of carbohydrates. The medium turns red on the slope and yellow on the bump, indicating that the microbe can only ferment glucose. TSI tests the ability of gram-negative rods. In addition to producing hydrogen sulfide (H2S), it is in charge of fermenting glucose, sucrose, and lactose. With a concentration of 10:10:1, sucrose, glucose, and lactose make up TSI. Approximately ten grams of sucrose, or 1%, are present in the medium, 10 portions of lactose, or 1%, and 1 portion of glucose, or 0.1%, is present. 65 grams of medium were mixed with one liter of distilled water in a flask. After thoroughly dissolving and mixing the agar, the medium was autoclaved for 15 minutes at 121°C. After autoclaving, the media was placed into glass tubes that had been sterilized. The tubes were susceptible to a 4-5 cm inclination. left to solidify in the media. The isolated colonies are removed from the solidified medium using the sterile straight inoculation needle. Streaking was done on the agar slant TSI's evident Stabbing was used to inoculate the agar from the middle of the medium to the test tube's bottom. The medium-filled tube was incubated at 35°C for 24 hours (Ahmad *et al.*, 2006).

Antimicrobial susceptibility test

The agar disc diffusion method using Mueller-Hinton Agar was used to determine the isolates' antibiotic sensitivity. Colistin sulfate, Gentamicine, Amoxicillin, Ciprofloxacine, and Enrofloxacine were the antibiotic discs (mg) that were utilized. The NCCLS of the minimum inhibitory concentration (MIC) break points and the reference zone diameter interpretative standard (millimeter) were used to evaluate the results. The strains will be classified as resistant, intermediate, and susceptible. Based on zones of inhibition and diameter, which were determined and compared with the zone size analysis chart provided by the source, the strain was categorized as susceptible (S), intermediate (I), and resistant (R) (Mengistu *et al.*, 2014).

Additionally, the multiple antibiotic resistance (MAR) index was calculated for each Shigella isolate using the method a/b, where "a" represents the amount of antimicrobials to which the isolate will be resistant and "b" represents the total amount of antimicrobials to which the isolate will be exposed. Mueller Hinton Agar is used to test for antibiotic susceptibility. The Bauer Kirby method now uses it as the standard and standards medium, and the NCCLS specifies how it should be carried out. One liter of distilled water was mixed with 38 grams of agar. To thoroughly dissolve it, heat the medium while stirring constantly, then boil it for one minute. The medium should be autoclaved at 121°C for 15 minutes. At room temperature, let it to cool. Now Fill sterile Petri plates with the prepared Mueller Hinton Agar. Petri dishes were left at room temperature to cool (Mengistu *et al.*, 2014).

Procedure

Mueller Hinton (MH) agar was used to test for antibiotic sensitivity. Autoclaved the media. The sterile petri plates were filled with media. The microbe was injected onto Mueller-Hinton agar after the medium had cooled. On the MH agar plates, several antibiotics (discussed above) were administered. Plates were stored in the incubator for a whole day. Zones of inhibition were measured using a ruler after a 24-hour period. This result was identified as sensitive and resistant to the specific drugs based on the zones of inhibition (Mengistu *et al.*, 2014).

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Results

In the present study, overall 200 liver samples of dead chicken were used to check the isolation identification and antibiogram of *shigella*. Liver samples were collected from broiler chicken from five age groups and from seven different areas of district Abbott bad. All these samples were examined by traditional method. After examination 36/200 sample were positive for *shigella* with incidence of 18% (Table 1).

Table 1: Incidence of Shigella in various samples

s.no	Source of sample	No of examined samples	No. of positive sample	No of negative samples	% of negative Samples	% of positive samples
1	Liver tissue	200	36	164	82%	18%

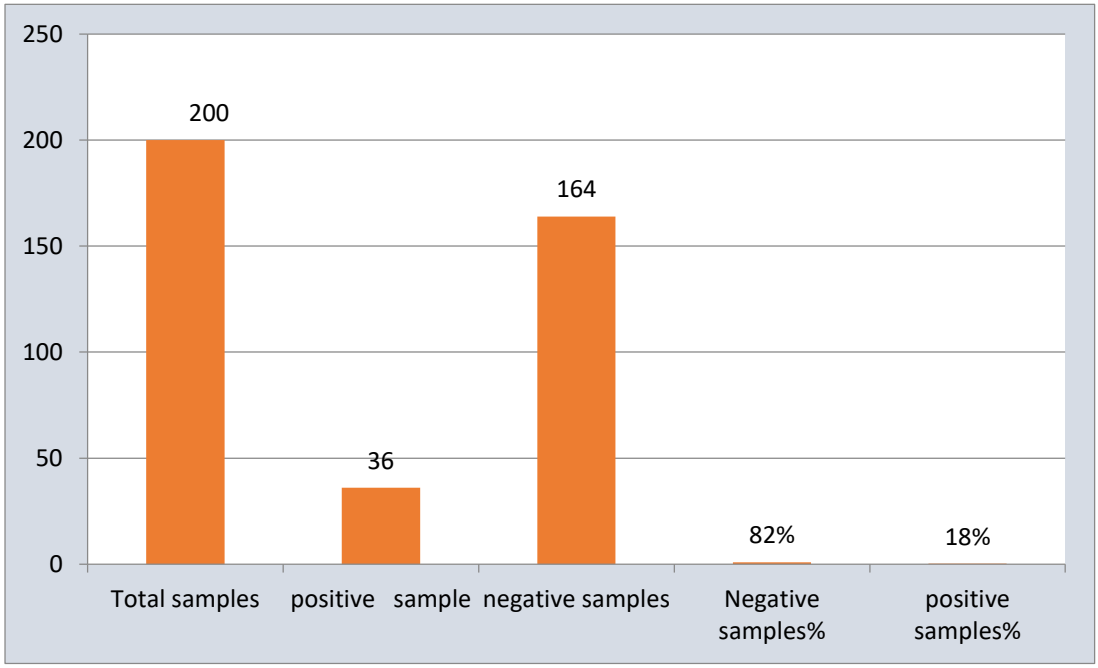

Figure 3: Incidence of shigella in various samples

Morphology of Shigella colonies

The growth on colonies exhibiting colorless appearance on SSA and green color on HEA media.as shown in table 2.

Table 2: Characteristic colony appearance on various culture media

S. No.	Media used	Colony appearance
1	SSA	Green colonies
2	*HEA*	Colorless colonies

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Colonies appearance on *HEA* agar

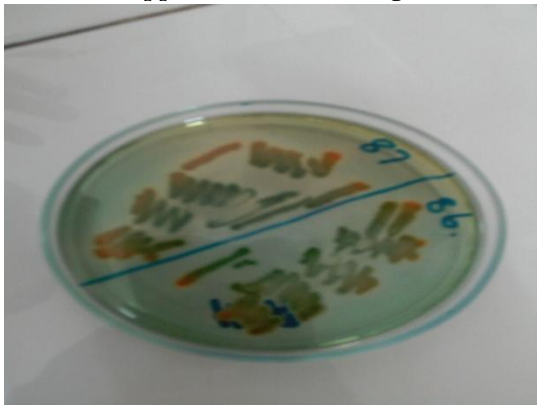

Figure 4

Colonies appearance on *SSA* agar

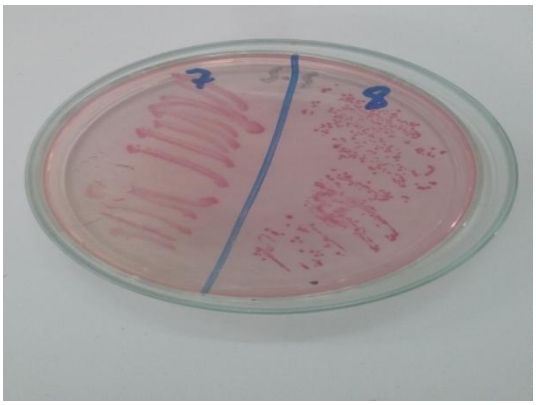

Figure 5

Biochemical test

Triple sugar iron test

Triple sugar iron agar (TSI) biochemical assays were shown to identify pure colonies. Figure 6 demonstrates that in the triple sugar iron test, isolates produced an acid butt and an alkaline slant without producing any hydrogen sulfide gas.

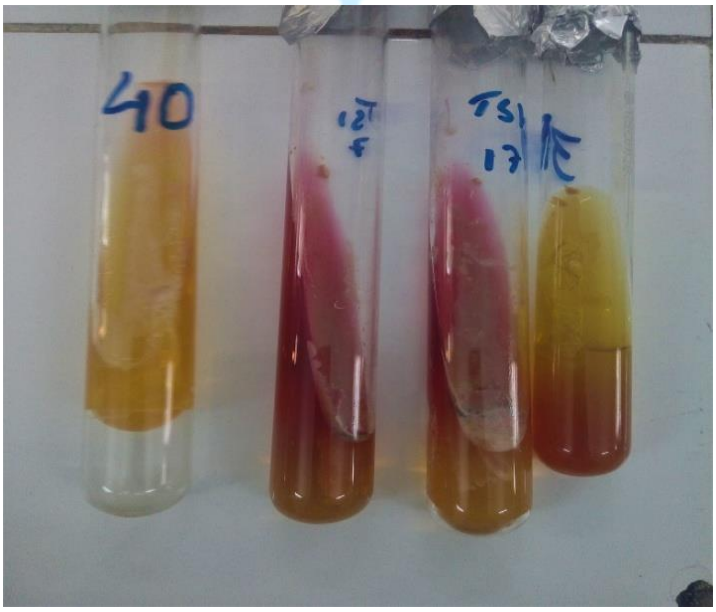

Figure 6: Culture tubes showing TSI slants

200 broiler samples collected from different areas of District Abbottabad shows different prevalence percentage as shown in table 3.

Table 3: Prevalence of *Shigella* spp. in broiler samples according to location

s.no	Location	No. of examined samples	No. of positive samples	No. of negative samples	% of positive samples	% of negative Samples

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1	Mangal	30	4	26	13%	86%
2	Nawasher	40	8	32	20%	89%
3	Havelian	22	8	14	36%	63%
4	Qalanderabad	32	4	28	12%	87%
5	Shimlapahari	28	2	26	7%	93%
6	Bandha ali khan	28	6	22	21%	74%
7	Salhad	20	4	16	20%	80%

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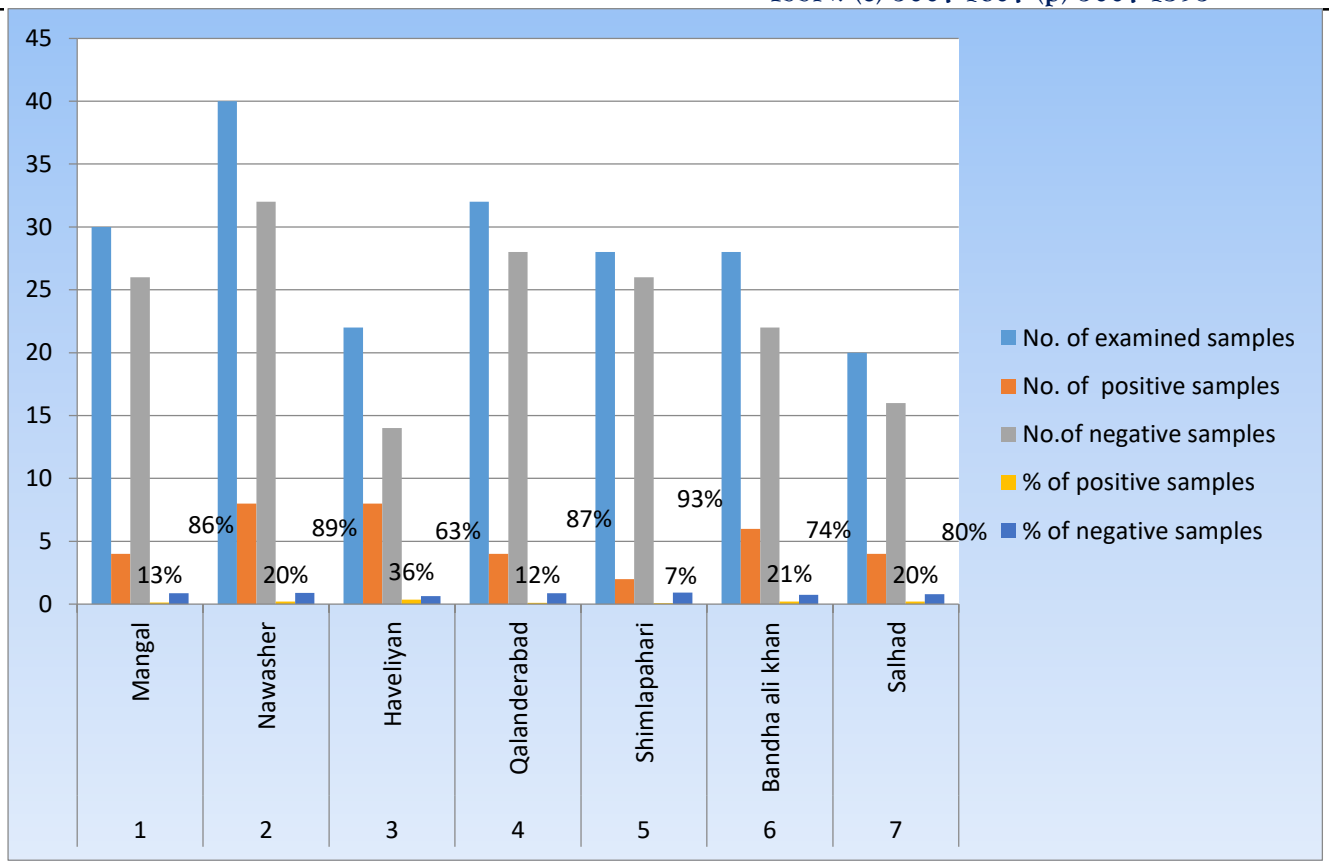

Figure 7: Prevalence of Shigella spp. in broiler samples according to location

Incidence of *Shigella* spp according to age

Samples were collected from different age-groups. After isolation, a high incidence was found in 8-14 days old broiler (25%) rather than 29 -35 days and 0-7 days (19%, 16%) respectively, and a low incidence 22-28 days 15-21 days and (11%.12%) was found.

Table 4: Incidence of *Shigella* spp. according to age

S.No	Age/days	No. of examined samples	No. of Positive samples	No. of Negative Samples	%of Negative samples	% of positive samples
1	0-7	36	6	30	83%	16%
2	8-14	56	14	42	75%	25%
3	15-21	48	6	42	88%	12%
4	22-28	18	2	16	89%	11%
5	29-35	42	8	34	81%	19%

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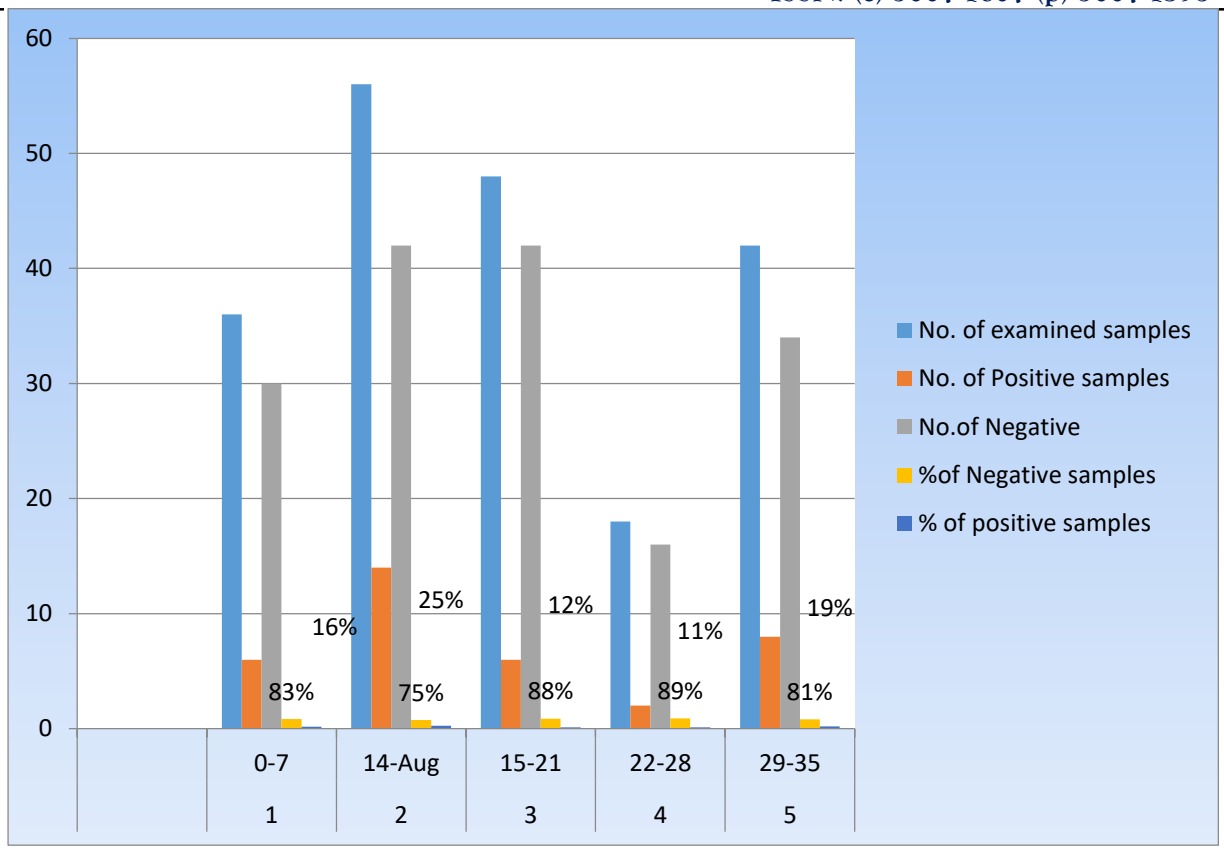

Figure 8: Incidence of Shigella spp. according to age

Table 5: Month wise prevalence of *Shigella* specie

S.NO	Month	Total number of samples	Positive samples	Negative samples	% of positive samples
1	Oct-17	30	10	20	33%
2	Nov-17	25	7	18	28%
3	Dec-17	36	2	34	6%
4	Jan-18	36	4	32	12%
5	Feb-18	40	4	36	10%
6	April-18	33	9	24	27%

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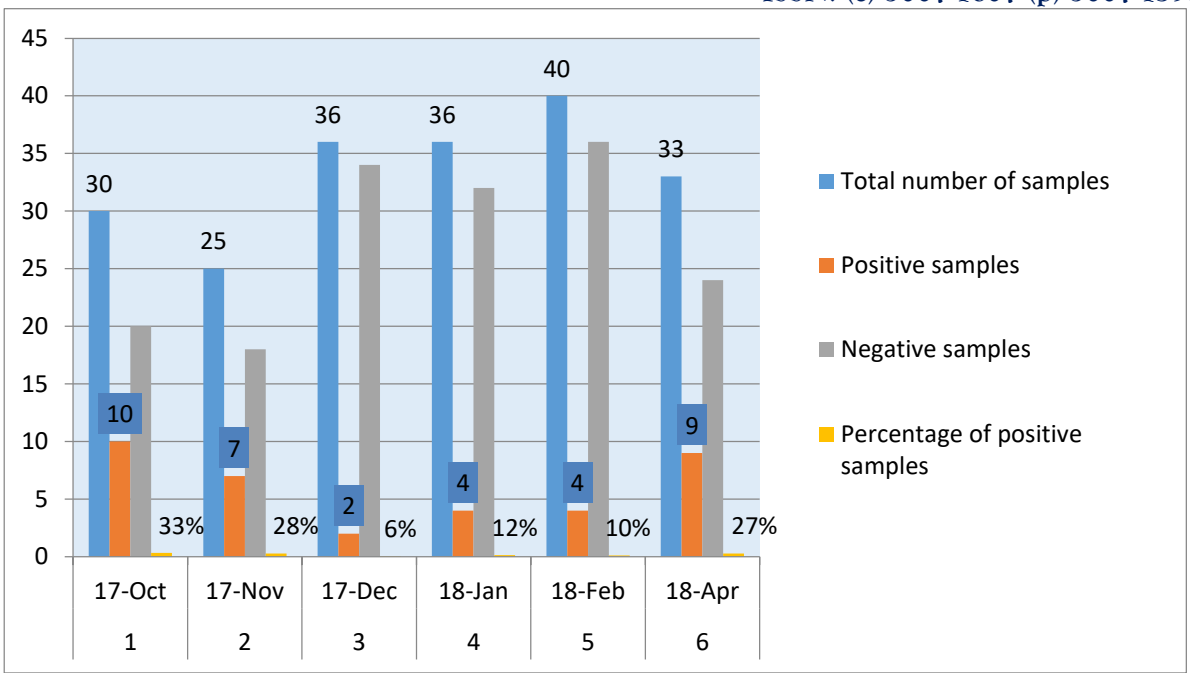

Figure 9: Month wise prevalence of *Shigella* specie

Using the disc diffusion method or Kirby-Bauer method, a total of 36 isolates were examined for antimicrobial drug resistance against five widely used antibiotics from various groups. Using five regularly given antibiotics, the disc diffusion technique was used to examine the antibiotic sensitivity and resistance patterns of isolated Shigella spp. Shigella shown varying degrees of resistance to different antimicrobial drugs. Gentamicin was shown to be the most sensitive antibiotic. Enrofloxacin and Colistin sulfate are intermediate, while Ciprofloxacin and Amoxicillin were determined to be the most resistant medications. Veterinarians would undoubtedly benefit from these findings when choosing the best medications to treat Shigella infections. The antimicrobial agent used with their disc concentration is mentioned in table 6

Table 6: Antimicrobial agents and their disc concentrations

S.NO	Antimicrobial agent	Symbol	Disc concentrations (µg/disc)
1	*Amoxicillin*	AML	10 µg
2	*Ciprofloxacin*	CIP	5µg
3	*Enrofloxacin*	ENF	5 µg
4	*Gentamicin*	CN	10 µg
5	*Colistin sulfate*	CS	10 µg

Legend: µg = Microgram

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Figure 10: Antimicrobial agents and their disc concentrations

Table 7: Comparison of standards with results

S no	Antibiotic	Disc concentrations (µg/disc)	Resistive	Intermediate sensitivity	Sensitive	average Results	Remarks	%
1	Amoxicillin	10 µg	13	14-17	18	2	Resistive	11.1
2	Ciprofloxacin	5µg	15	16-20	21	12.4	Resistive	68.9
3	Enrofloxacin	5 µg	16	17-22	23	14.3	Intermediate	90.5
4	Gentamicin	10 µg	12	13-14	14	16.9	Sensitive	82.8
5	Colistin sulfate	10 µg	10	15-18	19	15.9	Intermediate	88.3

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Discussion

The findings from the morphological analysis, culture, antimicrobial susceptibility, and biochemical examinations of the liver of deceased broiler chickens indicate that 36% of the samples were examined. Enterococci are responsible for the transmission of genes that confer resistance to antibiotics in the majority of animals' normal gut microbiota. Enterococci are the perfect bacterium for regulating factors associated with antibiotic susceptibility tests because of these characteristics. A considerable amount of morbidity and death is still caused by shigellosis, particularly in young chicks. In this investigation, the majority of shigella species were isolated from young chickens (Lampel *et al.,* 2018).

In the present study, no enrichment was used for the isolation of *Shigella* considering the work of (Warren *et al.,* 2006) who reported that the use of enrichment for the isolation of *shigella* were neither very specific nor sensitive. BAM (Bacteriological Analytical Manual) method also recorded that an effective selective enrichment procedure for *Shigella* is still not available (Cardoso, 2009).The suspected isolates of *Shigella* were primarily obtained by using XLD media, while SSA and HEA media were used for further confirmation. These findings agreed with the observations of Hunt (Talukder *et al.,* 2002) who recommended the parallel use of at least two plating media differing in selectivity. They proposed that the most effective culture medium for Shigella isolation was XLD agar. In the current investigation, Shigella was detected from feces using XLD agar in conjunction with MacConkey agar. Shigella species isolation using MLA, XLD, and SSA is also being done extensively (Nataro *et al.,* 2011).

From this study it was exposed that the total prevalence of *Shigella* were 36%, this result is an agreement with report of shigella from chickens has been documented in India by (Azmi et al., 2014). Okechuku *et al.,* (2018) reported 6.7% overall prevalence of *Shigella* in Nsuka Nigeria and the percentage of *Shigella* spp isolates were (3.2%), in 2004, first mentioned shigellosis in chicken defined by bloody and purulent dysentery in chickens (Yang *et al.,* 2007) that is less than our study, that is dissimilarity is due to geographic variation and changing patterns of serogroups and serotypes of shigella species from time to time. These variations in the general prevalence of Shigella might be caused by a number of issues, including the management system, environmental conditions, the chickens' resistance to Shigella, and the strains of chickens. Comparing the prevalence estimates derived from various research is challenging. They could indicate actual variations in Shigella dispersion across geographical areas and management systems, but they might also just be the result of variations in the methods employed to assess Shigella prevalence. These findings demonstrated that the apparent prevalence of Shigella varies according on sample types, detection methods, handling procedures, and collecting methods. These variations could obscure the effects of other elements, such seasonal trends and processing techniques, which are really altering the bacterial distribution and breeding methods.

The rise in antibiotics resistance had been reported in the past two decade (Kapil *et al.,* 2020) and antibiotic resistance still remains a global problem today. In intensively reared food animals, antibiotics are administered for therapeutic purpose and as Antimicrobial growth promoters (AMGPs) to the whole flock rather than individuals (Kapil *et al.,* 2020). The treatment of shigellosis may involve the use of antibiotics. While antibiotics are utilized, the most common ones used to treat shigellosis include ampicillin, trimethoprim, ciprofloxacin, nalidixic acid, and sulfamethoxazole. Shigella can be killed with routine therapy, but sadly, indiscriminate medication usage and horizontal gene transfer have generated Shigella species resistant to widely used antibiotics (Niyogi, 2005). The study indicated that Amoxicillin and Ciprofloxacin had high levels of antibiotic resistance (89% and 31%, respectively), while Enrofloxicin and Colistine Sulphate were found to be intermediate among the five antibiotics. Gentamicin, on the other hand, was shown to be sensitive (82.8%).

The susceptibility patterns to the antibiotics obtained in the present study for the *Shigella spp* is in agreement with the report of (Niyogi, 2005) that also reported high susceptibility to *gentamicin* but in variance with study of (Obi and Ike, 2018) who reported *gentamicin* resistive and with the study of (Obi and Ike, 2018) who reported

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60% sensitivity with respect to *ciprofloxacin*. Also (Ocean *et al.*, 2015) reported a high level of resistance to the newer *quinolones* including *ciprofloxacin* in *Shigella* isolates from poultry litterin Northern Nigeria. Shigella spp. isolates were 89% resistant to amoxicillin, according to antibiogram investigations, which is consistent with (Ocean *et al.*, 2015) study that found 100% resistance. The sensitivity pattern discrepancies between the current study and the previously stated studies may represent strain variance brought on by different sample sources.

Conclusion

Shigella is primarily found in early broiler chicks, according to the study mentioned above. It's clear that the infection spreads from the hatchery to the broiler chicks. By using non-contaminating feed and giving broilers an antiseptic environment, shigellosis risks may be reduced and egg contamination can be prevented. There will be a lower death rate than before if control measures are taken carefully. Shigellosis is a food poisoning disease that can be spread orally and produces bloody diarrhea in both humans and animals, particularly broiler chickens. It produces diarrhea that is bloody and pleurant. Using a variety of morphological, cultural, antimicrobial susceptibility, and biochemical assays, Shigella spp. were effectively identified and isolated from broilers in this investigation. The study was carried out in the Pakistani district of Abbottabad. Shigella spp. in broilers were shown to be multidrug resistant in the current investigation. Various Shigella species may exhibit differences. Due to the genetic diversity, Shigella spp. may become very resistant. The experiment's results also suggest that the use of gentamycin, choleric acid, and Enrofloxacin may be preferred in the clinical management of shigellosis, posing a danger to the chicken business. Future study should focus on molecular characterization and genomic investigations to better understand the genes causing the pathogenicity and treatment resistance of the Shigella isolates from broiler liver. These findings suggest that broilers may act as reservoirs or carriers for the spread of Shigella species, hence efforts should be taken to inform the local broiler-raising community about the potential public health risks.

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