Bulletin of Biological and Allied Sciences Research
***ISSN:* 2521-0092**
www.bbasr.org
DOI: https://doi.org/10.54112/bbasr.v2023i1.55
*Bull. Biol. All. Sci. Res., Volume, **8**: 55*

Original Research Article

ANTIBIOTIC SUSCEPTIBILITY PROFILE AND NUTRITIONAL EVALUATION OF RAW AND COOKED MEAT OF *CYPRINUS CARPIO*

MUHAMMAD Z[1], KHAN S[1*], KHATTAK F[2], BASHIR K[1], KHAN A[1], SHERAZ M[1], ULLAH I[1], KHAN MI[1], KHAN MA[1], ULLAH A*[1]

[1]*Department of Health and Biological Sciences, Abasyn University, Peshawar, 25000, Khyber Pakhtunkhwa Pakistan*
[2]*Center of biotechnology & microbiology (COBAM) University of Peshawar, Peshawar 25000, Pakistan*
**Correspondence author email address: aminbiotech7@gmail.com, sarakhan17301@gmail.com*

(Received, 4th February 2023, Revised 20th December 2023, Published 22nd December 2023)

Abstract *The Cyprinus carpio fish belongs to the family Cyprinidae and nutritionally consists of beneficial amino acids and omega-3 polyunsaturated fatty acids. However, consuming fish contaminated with pathogenic bacteria causes serious human health issues. The study aimed to identify pathogenic bacteria, antibiogram profile and extended-spectrum-beta-lactamase (ESBL) producing bacteria, and nutritional composition of C. carpio fish in Peshawar, Pakistan. Fish samples (30 uncooked and 30 cooked) were collected from Peshawar and analyzed in the laboratory to detect pathogenic bacteria. The bacteria were isolated, and different sorts of staining and biochemical tests including Catalase, Oxidase, Triple sugar iron, Urease, Citrate utilization, Indole etc,. were performed for identification. Antibiotic susceptibility pattern was tested on Muller Hinton agar medium (MHA) against nine antibiotics. Nutritional analysis of raw and cooked meat was performed using the Association of Official Analytical Chemists procedure. The results showed that E. coli has the highest resistance (100%) towards Tetracycline and Penicillin, while K. pneumonia showed the highest antibiotic resistance (100%) against Tetracycline, Oxacillin, Ampicillin, and Penicillin. Sheigilla was observed to the highest resistance against Penicillin and Oxacillin, which was 100%, and Salmonella showed the highest resistivity (100%) towards Tetracycline, Oxacillin, and Penicillin. Similarly, Tetracycline, Oxacillin, and Ampicillin showed the highest antibiotic resistance (100%) toward S. aureus. In 60 samples, 105 bacterial species (E. coli, Salmonella, Sheigella, Klebsiella, and S. aureus) were isolated. All isolated bacteria were ESBL producing except S. aureus. The results showed that crude fat (3.5-10.4%), crude fiber (0.23-0.38%), crude ash (5.6-5.7%), crude protein (69.03-89%), and moisture (4.6-5.8%) were greater in uncooked fish meat as compared to cooked fish meat. The study concludes that the high prevalence of pathogenic bacteria in uncooked meat is observed compared to cooked meat of C. carpio. Uncooked fish meat is more nutritious than cooked fish meat as the amount of protein, fats, fiber, ash, and moisture decreases during cooking.*

*[Citation: Muhammad, Z., Khan, S., Khattak, F., Bashir, K., Khan, A., Sheraz, M., Ullah, I., Khan, M.I., Khan, M.A., Ullah, A. (2023). Antibiotic susceptibility profile and nutritional evaluation of raw and cooked meat of cyprinus carpio. Bull. Biol. All. Sci. Res. **8**: 55. doi: https://doi.org/10.54112/bbasr.v2023i1.55]*
Keywords: *Cyprinus carpio; Cooked; Uncooked; Antibiotics; Pathogens; Extended-spectrum-beta-lactamase*

Introduction

In the kingdom, Animalia fish is the largest group, having a population of approximately 30,000 well-known species and used mainly for oil and food production (Seel *et al.*, 2016; Suseno *et al.*, 2014). Fish consumption as food is recommended because of its good digestibility and highly rich polyunsaturated fatty acids (PUFA). Freshwater fish is considered a nutritious food because it contains a high proportion of protein with a balanced amino acid profile, modest levels of healthy lipids, and a lower concentration of n-3 PUFA than marine fish. It also contains many vitamins and minerals like phosphorus, calcium, magnesium, sodium, and potassium (Raufu *et al.*, 2014; Obianime and Obire, 2017). Fish meat has a lower cholesterol content and is generally advised for consumption by the adult population (Ayeloja *et al.*, 2018). According to regulatory bodies, eating fish is recommended 1–2 days per week, with one part of oily fish containing 200–500mg of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Sobczak *et al.*, 2020). In the fish culture, health problems generally occur due to the interaction of host, pathogen, and environment (Salem *et al.*, 2020). A commodity can be divided into three categories: indigenous bacteria, nonindigenous bacteria (enteric bacteria), and contamination of fish by bacteria during storage, manufacturing or food preparation (Lerma-Fierro *et al.*, 2020). The fishery products are a major source and reservoir of pathogens that cause food

poisoning (Morshdy, 2014). Fish is considered a vector of pathogenic bacteria in humans, including a wide range of bacteria such as *Pseudomonas* sp., *Escherichia coli, Staphylococcus aureus, Vibrio* sp., *Mycobacterium* sp., *Salmonella* sp., *Shigella* sp., *Enterococcus* sp., and *Proteus* species etc. (Suseno *et al*., 2014). Salmonella and Shigella species are found commonly in fish and other aquatic organisms (Alkhunni et al., 2017) and cause major public health problems (Lamboro *et al.,* 2016).

In the last few decades, aquaculture production has shown a rapid growth for the world's growing population. However, aquatic organisms are the main reservoir of pathogens and harm human health and aquatic organisms. For this reason, careful monitoring and evaluation of foodstuffs in terms of human health is necessary. The microbes of fresh and processed aquatic organisms should be examined carefully in the evaluation stage. The recent study aims to screen the microbial pathogens in raw and cooked meat of *Cyprinus carpio* and compare the nutritional composition (crude fat, protein, carbohydrate, ash, and fibers) of raw and cooked fish meat.

Materials and methods

Sample Collection

The study was conducted at the Microbiology Laboratory, Department of Health and Biological Sciences, Abasyn University Peshawar, from October 2021 to February 2022. From different local markets in Peshawar, 60 samples of *C. carpio* (30 cooked and 30 uncooked fish) were collected, mixed with soft ice, put in an icebox, and transported to the Department of Health and Biological Sciences, Abasyn University Peshawar. The *C. carpio* was identified by observing special characteristics. After collection, the samples were kept at room temperature and used within 24hrs for experimental purpose.

Fish Sampling

Peptone water was prepared to make serial dilutions. Peptone water was sterilized by autoclaving at 15 lbs. pressure (121°C) for 15 minutes and then cooled. Peptone water (9ml) was added to test tubes. Then 1 gram of fish flesh and 10 ml peptone water were taken in another tube and carefully shaken. 1 ml of processed fish flesh was added to the first test tube containing 9 ml of peptone water. This first tube was labeled as 10-1. Similarly, 1 ml of the first test tube was mixed with 9 ml of peptone water in another test tube labeled 10-2. In this way, dilutions up to 10-9 were prepared.

Isolation of Bacteria

To isolate pathogenic bacteria, dilution numbers 10-3, 10-4, 10-5, 10-6, and 10-7 were used. From each dilution, 100 µl sample was taken and spread plated onto different selective media i.e. MacConkey agar plates, *Salmonella Shigella (SS)* agar plates, and Mannitol salt agar (MSA) plates. The plates were dried and then placed in an incubator in an inverted position overnight at 37 °C. For *E. coli* and *Klebsiella pneumonia* confirmation, dark pink colonies were chosen and streaked on new McConkey agar plates. Transparent black dot colonies were chosen and streaked on fresh S-S agar plates for confirmation of *Shigella* and *Salmonella*. Yellow colonies were chosen and streaked on fresh MSA plates for the confirmation of *S. aureus*.

Identification of Isolates

Colonies of bacterial pathogens were identified by examining size, shape, pigmentation, morphology, and biochemical characterization (Alameer *et al.*, 2020). Different sorts of staining and biochemical tests, including Catalase, Oxidase, Triple sugar iron, Urease, Citrate utilization, Indole, Coagulase, and Hydrogen sulfide production tests, were performed for the identification of gram-negative bacteria using standard methods (Tsfaye *et al.,* 2018). Extended-spectrum beta lactamase-producing bacterial isolates were identified by standard techniques using Muller Hinton agar media (MHA).

Antibiotic sensitivity testing

For antibiotic susceptibility discs, the diffusion method was performed using MHA media. The antibiotics Tetracycline (10µg, Oxoid), Cefotaxime (30µg Oxoid), Penicillin (10µg, Oxoid), Oxacillin (1µg, Oxoid), Amikacin (30µg, Oxoid,), Streptomycin (10µg, Oxoid), Ampicillin (10µg, Oxoid), Chloramphenicol (30µg, Oxoid) (10µg, Oxoid), and Erythromycin (15µg, Oxoid) were used. Zones of inhibition were measured after incubation. The resistance levels were explained as described by the Clinical and Laboratory Standards Institute (CLSI, 2010) (Pamuk *et al.,* 2019).

Nutritional analysis

Nutritional analysis of raw and cooked meat was performed using the Association of Official Analytical Chemists procedure (Tobaruela *et al.,* 2018). The study was conducted at the livestock Research and Development department, Veterinary Research Institute (VRI) Peshawar, Pakistan. Samples were dried in an oven at 105°C for 24h. The ash content was determined after incineration at 550°C for 6 hours. This method measured the composition of other components, such as crude fat, dry matter, crude protein, and fibers (Sobczak *et al*., 2020).

Statistical Analysis

The statistical analysis was performed using MS Excel and GraphPad Prism software. All the data and graphs, including the tables, were statistically analyzed, and their values were calculated accurately.

Results

60 common carp (C. carpio) samples were taken, i.e., 30 cooked and 30 uncooked fish samples. Different bacteria were isolated and identified based on colony morphology, gram staining, and biochemical test. Bacterial pathogens were isolated from different samples of *C. carpio*. The bacterial isolates were *S. aureus, Salmonella, Shigella, E. coli,* and *Klebsiella* in cooked and uncooked fish. The results indicate that uncooked fish were more contaminated with bacterial pathogens than cooked fish (Figure. 1).

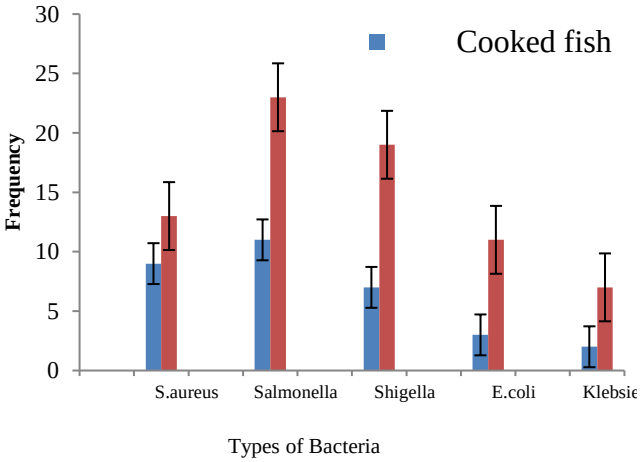

Figure 1: Frequency distribution of bacteria in cooked and uncooked fish

Colony Morphology

The bacterial isolates were identified based on colony Morphology using MacConkey, Salmonella-Shigella (S-S), and Mannitol Salt Agar (MSA) Media. All E. coli and Klebsiella colonies were dark pink with a halo of bile precipitate surrounding them, suggesting vigorous lactose fermentation. All the colonies of *Shigella* were clear, colorless, and transparent, while colonies of *Salmonella* were colorless, clear, and transparent with black holes by producing H_2S gas observed on S-S agar media. All the colonies observed on MSA media were yellow, as shown in figure 2.

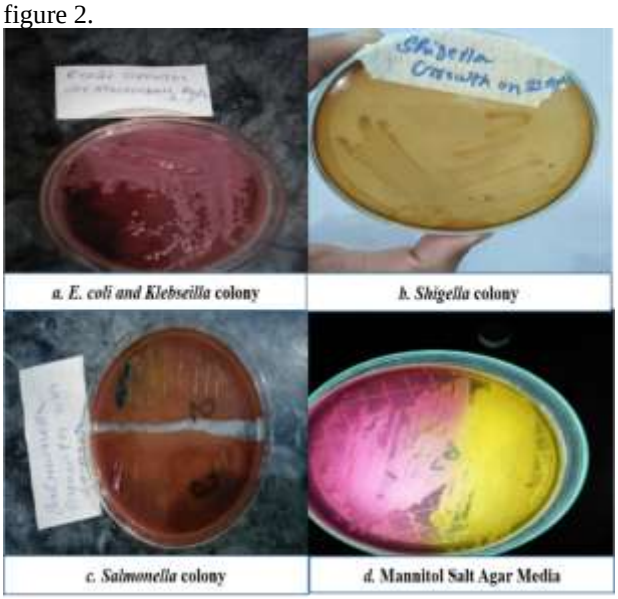

Figure 2: Growth of *E. coli* and *Klebsiella, Shigella,* and *Salmonella* and *S. aureus* (MSA media)

Gram Staining

All isolates (*Salmonella, Shigella, E. coli, and Klebsiella*) appeared rod-shaped and pink-colored under the microscope, while *S. aureus* appeared cocci-shaped and purple-colored under the microscope (Figure 3).

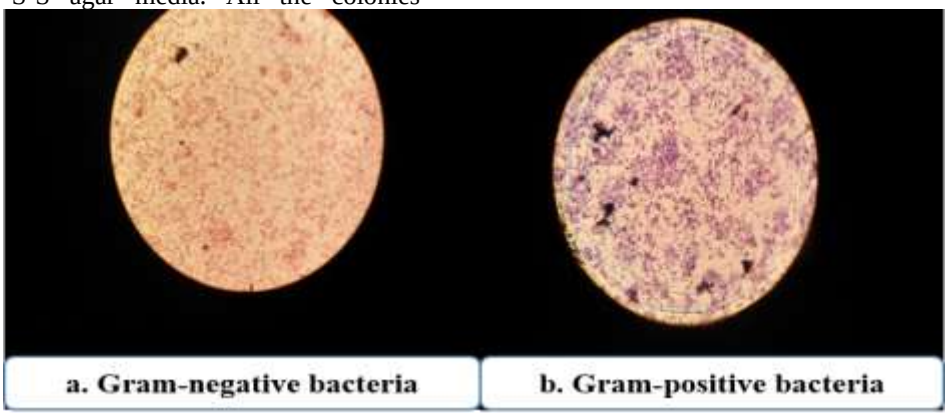

Figure. 3: Gram staining of isolated colonies under microscope (a) and (b)

Biochemical Tests Results for Bacterial isolates

Biochemical tests were performed for bacterial isolates (E. coli, Salmonella, Shigella, Klebsiella, S. aureus) in both cooked and uncooked fish meat, as shown in table 1. The complete biochemical test results of microbes were shown in the supplementary data (Table 1a)

Table. 1 Frequency of bacterial isolates identified in cooked and uncooked fish meat

Bacterial isolates	No of isolate	Urease test	Citrate test	Indole production test	Oxidase test	Triple Sugar Iron test			Catalase test	Coagulase test
						Butt/ Slope	Gas	H_2S		
E. coli	14	-	-	+	-	Acidic/ acidic	+	-	+	-
Klebsiella	9	+	+	-	-	Acidic/ acidic	+	-	+	-
Shigella	26	-	-	-	-	butt / acidic	-	-	-	-
Salmonella	34	-	+	-	-	butt/acidic	+	+	-	-
S. aureus	22	+	-	+	-	Acidic /acidic	-	-	+	+

Symbol: (–) negative and (+) positive

Detection of Extended Spectrum Beta-Lactamase (ESBL) producing bacteria

he bacterial isolates in cooked and uncooked fish meat were detected for ESBL. Salmonella, Shigella, E. coli, and Klebsiella were out of these bacterial isolates and revealed positive results, determining that these bacterial strains contain beta-lactamase enzymes. These bacterial strains were resistant to some antibiotics i.e. Clavulanic acid and Ceftazidime. The results of ESBL-positive bacteria are shown in figure 4.

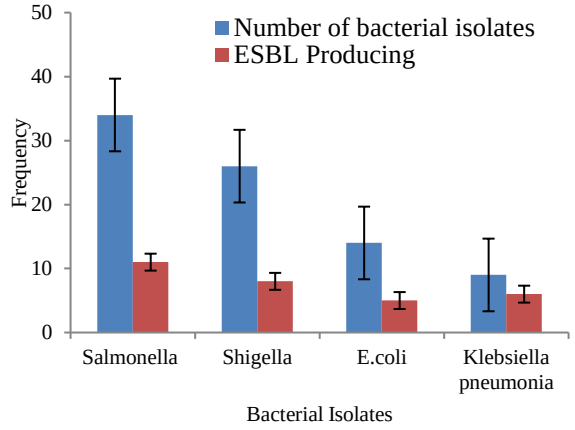

Figure 4: Frequency of ESBL-producing bacteria in cooked and uncooked fish

Susceptibility pattern of isolated *E. coli*

The resistivity and sensitivity to nine commercially available antibiotic or antibacterial agents were performed by disc diffusion method (Figure. 5). The study observed that *E. coli* showed the highest resistance (100%) and 0% sensitivity towards Tetracycline and Penicillin, while the least resistance was observed towards Streptomycin (7.1%) with 71.4% sensitivity. The results disclosed resistance towards other antibiotics such as Cefotaxime (21.4%), Oxacillin (85.7%), Ampicillin (85.7%), Chloramphenicol (92.8%), Penicillin (100%), Erythromycin (14.8%) and Amikacin (78.5%). The sensitivity of *E. coli* towards these antibiotics Cefotaxime, Oxacillin, Ampicillin, Chloramphenicol, Erythromycin, and Amikacin, were 78.5, 14.2, 0, 7.1, 85.7, and 21.4, respectively. Results are summarized in the figure 6.

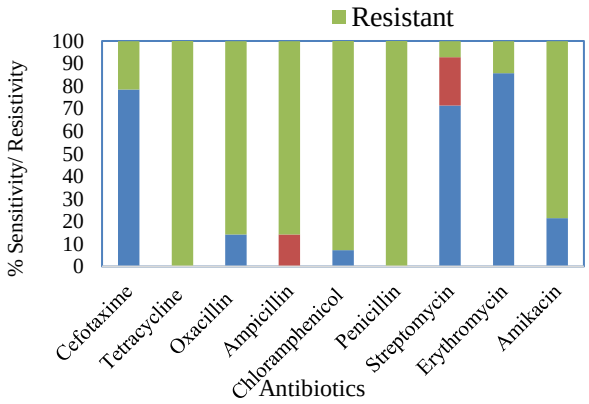

Figure 5: Antibiotics Subsceptibility Test

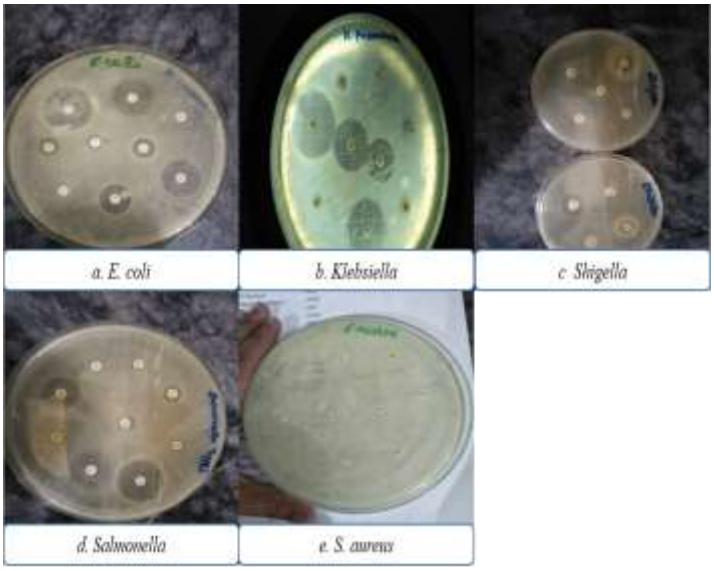

Figure 6: Susceptibility Pattern of *E. coli* towards Antibiotics

Susceptibility pattern of isolated bacteria *K. pneumoniae*

The susceptibility test for *K. pneumoniae* towards different antibiotics was reported. Tetracycline, Oxacillin, Ampicillin, and Penicillin showed the highest antibiotic resistance (100%) against *K. pneumoniae* with the least sensitivity 0%. The resistance. The resistance of *K. pneumoniae* towards these antibiotics Cefotaxime, Chloramphenicol, Streptomycin, Erythromycin, and Amikacin were 0, 33.3, 22.2, 66.6, and 33.3%, respectively. The highest sensitivity was observed towards Cefotaxime (88.8%), followed by Streptomycin (77.7%), Chloramphenicol (66.6%), Amikacin (66.6%) and Erythromycin (3.3%) as given in the Figure 7.

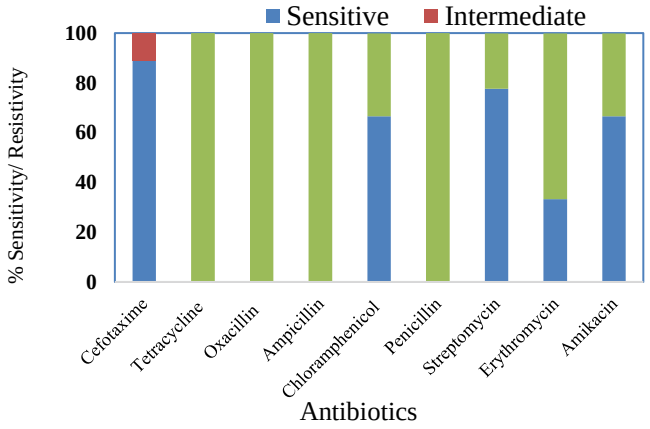

Figure 7: Susceptibility Pattern of *K. pneumoniae* towards Antibiotics

Susceptibility pattern of isolated *Shigella*

Shigella's sensitivity and resistance pattern were tested against selected antibiotics; the highest resistance was observed against Penicillin and Oxacillin, which is 100%, while 0% sensitivity was observed. The result showed the highest sensitivity of *Shigella* to Streptomycin (88.4%), followed by Amikacin (80.7%), Tetracycline (23.1%), Cefotaxime

(7.6%) and Erythromycin (7.6%). The resistance Cefotaxime, Tetracycline, Ampicillin, Chloramphenicol, Streptomycin, Erythromycin, and Amikacin noted was 92.3, 76.9, 9.3, 92.3, 11.5, 76.9 and 7.6%, respectively as shown in the Figure 8.

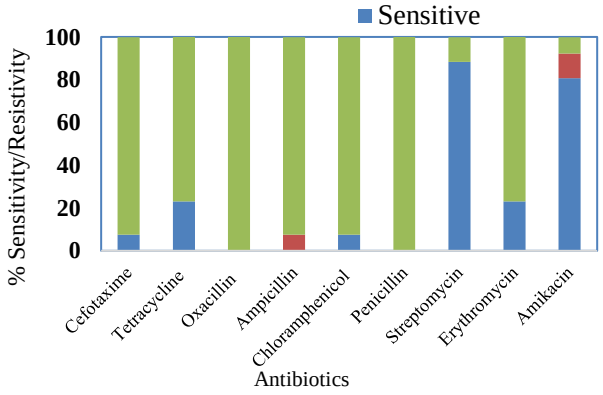

Figure 8: Susceptibility Pattern of *Shigella* towards Antibiotics

Susceptibility pattern of isolated *Salmonella*

In the study, antibiotics were tested for susceptibility of *Salmonella* for resistivity and sensitivity. Salmonella showed the highest resistivity (100%) and 0% sensitivity towards Tetracycline, Oxacillin, and Penicillin, while the least resistance was observed towards Amikacin (5.8%). The results showed that *Salmonella* was resistant to other antibiotics such as Cefotaxime (11.7%), Ampicillin (82.3%), Chloramphenicol (67.6%), Streptomycin (88.2%), Erythromycin (91.1%) and Amikacin (5.8%). The sensitivity of *Salmonella* towards Cefotaxime, Ampicillin, Chloramphenicol, Streptomycin, Erythromycin, and Amikacin were 88.4, 5.8, 32.3, 11.7, 8.8 and 88.2%, respectively (Figure 9).

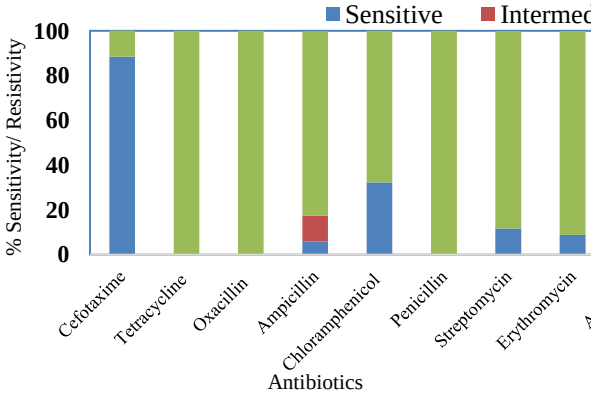

Figure. 9 Susceptibility Pattern of *Salmonella* towards Antibiotics

Susceptibility pattern of isolated bacteria *S. aureus*

The susceptibility test for *S. aureus* towards different antibiotics was reported. In the study, *Tetracycline*, *Oxacillin*, and Ampicillin showed the highest antibiotic resistance (100%) against *S. aureus* with the least sensitivity (0%). The resistance of *S. aureus* towards Cefotaxime, Chloramphenicol, Streptomycin, Erythromycin, and Amikacin were 4.5, 86.3, 81.8, 90.9 and 86.3%, respectively. The highest sensitivity was observed towards Cefotaxime (95.5%), followed by Streptomycin (18.1%), Chloramphenicol (13.6%), Amikacin (13.6%), and Erythromycin (9.1%) as given in figure 10.

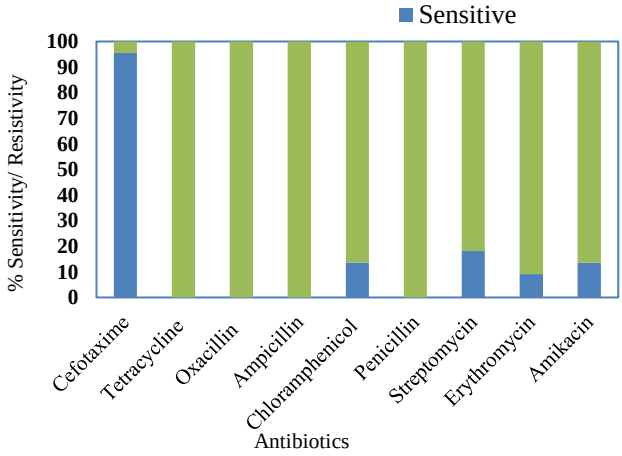

Figure 10: Susceptibility Pattern of *S. aureus* towards Antibiotics

Nutritional Analysis of Cooked and Raw Fish

The parameters determined for uncooked (Table 2) and cooked fish (Table 3) showed that the amount of fat, fiber, ash, protein, and moisture was higher in uncooked fish. After cooking these fish, the amount of these parameters decreased.

Table 2: Composition of uncooked fish

Uncooked fish Sample	% Dry matter (Fresh)	% Dry Matter (Ground)	On Dry Matter Basis				
			% Crude Fat	% Crude Fiber	% Crude Ash	% Crude Protein	% Moisture
S	20.68	94.20	3.475	0.23	5.69	89.00	5.80
Fish sample 2	20.25	95.26	8.27	0.355	5.745	85.78	4.74
Fish sample 3	21.90	95.35	10.39	0.38	5.56	69.03	4.65

Table 3: Composition of cooked fish

Cooked fish Sample	% Dry Matter (Fresh)	% Dry Matter (Ground)	On Dry Matter Basis				
			% Crude Fat	% Crude Fiber	% Crude Ash	% Crude Protein	% Moisture

Fish sample1	17.85	90.50	2.47	0.17	3.24	83.14	4.14
Fish sample 2	18.67	91.26	6.57	0.257	3.455	84.78	3.24
Fish sample 3	19.36	92.25	9.89	0.27	4.76	63.03	3.66

Discussion

The *C. carpio* fish is also one of the most common edible fish, an important source of protein and omega 3 fatty acids. However, *C. carpio*, especially in an uncooked form containing bacterial pathogens resistant to antibiotics, can be transmitted to consumers, causing various diseases. Similarly, cooked *C. carpio* contains fewer bacterial pathogens than uncooked fish meat. Antibiotic-resistant bacteria isolated from fish are quite common, and these antibiotic-resistant, beta-lactamase-producing food-borne bacteria might be transferred to food handlers and consumers. Uncooked fish have high nutritional content but more contamination of bacterial pathogens, while cooked fish meat contains fewer bacterial pathogens and less nutritional value (Seel *et al*.**,** 2016). In this study, bacterial pathogens such as *E. coli*, *Klebsiella*, *Salmonella*, *Shigella*, and *Staphylococcus aureus* were isolated and identified from uncooked *C. carpio* in Pakistan while in cooked fish the bacterial pathogens were *Salmonella*, *Shigella*, *S. aureus* and few *Klebsiella* and *E. coli*. The high prevalence of these bacterial pathogens in uncooked fish compared to cooked fish is consistent with the findings of Suseno *et al*. (2014). In another study, we observed different biochemical tests for bacterial confirmation. The Urease test showed that *E. coli*, *Salmonella*, and *Shigella* isolates were negative, while *Klebsiella pneumonia* and *Staphylococcus aureus* isolates were positive for the test. Citrate tests performed for these bacteria showed that *E. coli*, *Staphylococcus aureus*, and *Shigella* isolates were negative, while *Klebsiella pneumonia* and *Salmonella* isolates were positive for the test. Indole production test showed that Klebsiella pneumonia, Salmonella and Shigella isolates were negative while positive E. coli and Staphylococcus aureus isolates were reported. Oxidase test performed for all bacterial isolates (*E. coli, Salmonella, Shigella, Klebsiella, and S. aureus*) were negative. Similarly, on Triple sugar iron (TSI) test, bacterial isolates of *E. coli* and *Klebsiella* were positive, and *Shigella* were positive for this test but negative for gas and H_2S production, *Salmonella* was positive for this test, producing gas and H_2S, *S. aureus* was positive but did not produce gas and H_2S. Catalase test performed for *E. coli, Klebsiella, and S. aureus* were positive and negative for *Salmonella, Shigella*. Coagulase test was performed for these bacteria, which showed that *E. coli, Klebsiella pneumonia, Shigella*, and *Salmonella* were negative *Staphylococcus aureus* isolates were positive for this test. Our results are similar to the previous research study of Kousar *et al*. (2019).

Another finding of our study is the bacterial isolates in cooked and uncooked fish meat were detected for Extended Spectrum Beta- Lactamase (ESBL) production. The Enterobacteriaceae *Salmonella, Shigella, E. coli*, and *Klebsiella* showed positive results for extended-spectrum beta-lactamase (ESBL) enzymes. This report is similar to the findings of Alkhunni *et al*. (2017), who determined the presence of beta-lactamase enzymes that inactivate beta-lactam antibiotics. Here we observed that *E. coli* was highly resistant to Tetracycline and Penecillin and was highly sensitive to Ampicillin, Streptomycin, and Amikacin. Our results are similar to the findings of Shahriar *et al*. (2019), who found that E. coli is highly resistant to tetracycline and penicillin. In our study, the *Klebsiella* was resistant to Tetracycline, Oxacillin, Ampicillin, and Penicillin and were highly sensitive to Cefatoxime. The results of the present study are similar to the previous study reported by Das *et al*. (2018). Shigella's susceptibility patterns agreed with the research findings of Alkhunni et al. (2017). *Salmonella* isolates were also tested for susceptibility patterns, and the results are closed with the study of Lunested *et al*. (2015). In the study, nutritional analysis for uncooked and cooked fish revealed that the amount of fat, fiber, ash, protein, and moisture was higher in uncooked fish than in cooked fish. The percentages of crude fat in cooked fish were less than in uncooked fish. This report is similar to the findings of Sobczaka *et al*. (2020). The moisture percentage was also less in cooked fish than in uncooked fish. The results of the present study are consistent with those of Chakroborty *et al*. (2017). The percentage of crude protein in uncooked fish was less in cooked fish than in uncooked fish. The research study's findings are comparable with the results reported previously by Ivanova *et al*. (2018), in which the protein amount in cooked fish was more than in raw fish.

Conclusion

The prevalence of these bacterial pathogens and the nutritional study of *C. carpio* has been reported for the first time in Pakistan. The study concludes that uncooked fish meat is more nutritious than cooked fish meat as the amount of protein, fats, fiber, ash, and moisture decreases during cooking. A high frequency of pathogenic bacteria is observed in uncooked meat compared to cooked meat of *C. carpio* fish. From the study, most pathogenic bacteria completely resist antibiotics, and this can lead to big issues for the population that is mostly fond of eating fish. The fish can be a transmission source for antibiotic-resistant S. *aureus*, *Salmonella*, *Shigella*, *Klebsiella*, and *E. coli*, which can directly or indirectly infect people by consuming fish flesh (greater in raw fish than cooked fish). The Prevalence of bacterial pathogens and Antibiotic susceptibility data in Pakistan is quite alarming. These bacteria may colonize the human

gastrointestinal system, producing severe illnesses including Salmonellosis, Shigellosis, and dysentery, among other things, posing a serious health risk.

References

Alameer, A. H., Atshan, O. F., Mahmood, M. M., Al-Jewari, W. M., & Mohammed, A. A.(2020).Detection of *Salmonella* Species In Viscera Of Carp Fish.

Alkhunni, S. B. A., Gaballah, M. S. M., &Gultepe, N. (2017). Pathogenic bacteria for human and fish isolated from fish farm in Kastamonu, Turkey. *Journal of aquaculture and marine biology*, 6(3), 00157. DOI: 10.15406/jamb.2017.06.00157

Ayeloja, A. A., George, F. O. A., Jimoh, W. A., Shittu, M. O., &Abdulsalami, S. A. (2018). Microbial load on smoked fish commonly traded in Ibadan, Oyo State, Nigeria. *Journal of Applied Sciences and Environmental Management*, 22(4), 493-497.7 DOI: 10.4314/jasem.v22i4.9

Chakroborty, T., & Chakraborty, C. S. (2017). Comparative Analysis of nutritional composition and microbial quality of salt-smoke-dried mirror carp (*Cyprinus carpio* var. *Specularis*) during storage at 22-28 C and 4 C. *International Journal of Food Science and Nutrition*, *1*, 86-89.

Das, A., Acharya, S., Behera, B. K., Paria, P., Bhowmick, S., Parida, P. K., & Das, B. K. (2018). Isolation, identification and characterization of *Klebsiella pneumoniae* from infected farmed Indian Major Carp *Labeo rohita* (Hamilton 1822) in West Bengal, India. *Aquaculture*, *482*, 111-116. DOI: https://doi.org/10.1016/j.aquaculture.2017.08.037

Ivanova, A., Angelova-Romova, M., Petkova, Z., Antova, G., & Hubenova, T. (2018). Chemical composition and fatty acid profile of lipids in carp (*Cyprinus carpio* L.) meat as affected by cooking methods. *Bulgarian Journal of Agricultural Science*, *24*, 2.

Kousar, R., Shafi, N., Andleeb, S., Ali, N. M., Akhtar, T., & Khalid, S. (2019). Avaliação e incidência de patógenosbacterianosassociados a peixesemincubadoras da Caxemira de Azad, Paquistão. *Brazilian Journal of Biology*, 80(3), 607-614. DOI: https://doi.org/10.1590/1519-6984.217435

Lamboro, T., Ketema, T., &Bacha, K. (2016). Prevalence and antimicrobial resistance in *Salmonella* and *Shigella* species isolated from outpatients, Jimma University Specialized Hospital, Southwest Ethiopia. *Canadian Journal of Infectious Diseases and Medical Microbiology*. DOI: https://doi.org/10.1155/2016/4210760

Lerma-Fierro, A. G., Flores-López, M. K., Guzmán-Robles, M. L., & Cortés-Sánchez, A. D. J. (2020). Microbiological evaluation of minimally processed and marketed fish in popular market of the city of Tepic Nayarit, Mexico Sanitary quality of tilapia (*Oreochromis niloticus*). *Tropicultura*.

Lunestad, B. T., Nesse, L., Lassen, J., Svihus, B., Nesbakken, T., Fossum, K., & Yazdankhah, S. (2007). *Salmonella* in fish feed; occurrence and implications for fish and human health in Norway. *Aquaculture*, *265*(1-4), 1-8. DOI: https://doi.org/10.1016/j.aquaculture.2007.02.011

Morshdy, A. E. D. (2014). Prevalence of Antibiotic Resistant *Salmonella* in Fish and Shell Fish in El Sharkia Governorate Alaa El-Dien Morshdy, Rasha Mohammmed El Bayomi, Aya E. Abo Elfadl.

Obianime N. E & Obire, O. (2017).Microbiological and physico-chemical characteristics of fish ponds in Port Harcourt. *Science and Technology*, 4(1), 126-139.

Pamuk, S., İnat, G., & Sırıken, B. (2019).Prevalence, serotypes distribution and characterization of *Salmonella* in common carp (*Cyprinus Carpio*), Afyonkarahisar Province, Turkey. 18(4), 924-940. DOI: 10.22092/ijfs.2018.117833

Raufu, I. A., Lawan, F. A., Bello, H. S., Musa, A. S., Ameh, J. A., & Ambali, A. G. (2014). Occurrence and antimicrobial susceptibility profiles of *Salmonella serovars* from fish in Maiduguri, sub-Saharah, Nigeria. *The Egyptian Journal of Aquatic Research*, *40*(1), 59-63. DOI: https://doi.org/10.1016/j.ejar.2014.01.003

Salem, M., Zharan, E., Saad, R., & Zaki, V. (2020). Prevalence, molecular Characterization, virulo-typing, and antibiotic resistance of motile aeromonads isolated from Nile tilapia farms at northern Egypt. *Mansoura Veterinary Medical Journal*, *21*(1), 56-67. DOI: 10.21608/MVMJ.2020.21.108

Seel, S. K., Kabir, S. M. L., & Islam, M. A. (2016). Molecular detection and characterization of *Salmonella* spp. isolated from fresh fishes sold in selected upazila markets of Bangladesh. *Bangladesh Journal of Veterinary Medicine*, *14*(2).

Shahriar, A., Akter, T., Kobra, A. T., Emran, T. B., Mallick, J., & Dutta, M. (2019). Isolation of Pathogenic and Non-pathogenic Microbial Stains from Different Types of Sea Fish Samples and their Quality Assessment with Antibiogram Properties. *Journal of Adv. in Microbio*, 1-10. DOI: 10.9734/JAMB/2019/v19i130185

Sobczak, M., Panicz, R., Eljasik, P., Sadowski, J., Tórz, A., Żochowska-Kujawska, J. & Dias, J. (2020). Quality improvement of common carp (*Cyprinus carpio* L.) meat fortified with n-3 PUFA. *Food and Chemical Toxicology*, *139*, 111261. DOI: https://doi.org/10.1016/j.fct.2020.111261

Suseno, S. H., Hayati, S., & Izaki, A. F. (2014). Fatty acid composition of some potential fish oil from production centers in Indonesia. Oriental Journal of Chemistry, 30(3), 975-980. DOI: http://dx.doi.org/10.13005/ojc/300308

Tobaruela, E. D. C., Santos, A. D. O., de Almeida-Muradian, L. B., Araujo, E. D. S., Lajolo, F. M., & Menezes, E. W. (2018). Application of dietary fiber method AOAC 2011.25 in fruit and comparison with AOAC 991.43 method. *Food chemistry*, *238*, 87-93. DOI: https://doi.org/10.1016/j.foodchem.2016.12.068

Tsfaye, S., Kasye, M., Chane, M., Bogale, B., & Abebeagre, Z. (2018). Preliminary Survey of Gram-Negative Bacterial Pathogens from Commonly Caught Fish Species (*Oreochromis niloticus*, *Cyprinus carpio* and *Clarias gariepinus*) in Lake Hayiq, Ethiopia. *Fish Aqua J*, *9*(238), 2. DOI: 10.4172/2150-3508.1000238

Declarations

Conflict of Interest

The authors have declared no conflict of interest

Declaration of Interest Statement

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Acknowledgments

Not applicable

Author's contributions

ZM conducted research and wrote initial draft of manuscript. SK, FK, KB, and AU collected the literature and wrote the manuscript. AK, MS, IU, MI K, and MAK edit the manuscript in original. All authors have read and approved the final manuscript.

Ethics approval and consent to participate

Not applicable

Consent for Publication

Not applicable