A seminar paper

On

Antibiotics Toxicity in Fish and Its Impact on Public Health

<u>Submitted to</u>

Course Instructors	Major Professor
Dr. A. K. M. Aminul Islam Professor, Dept. of Genetics and Plant Breeding, BSMRAU	**Dr. Farhana Haque** Associate Professor, Dept. of Fisheries Management, BSMRAU
Dr. Satya Ranjan Saha Professor, Department of Agroforestry and Environment, BSMRAU	
Dr. Shaikh Shamim Hasan Professor, Dept. of Agricultural Extension and Rural Development, BSMRAU	
Dr. Dinesh Chandra Shaha Associate Professor, Dept. of Fisheries Management. BSMRAU	

<u>Submitted by</u>

Anasua Kar

Reg. no. 17-05-4288

MS student

Dept. of Fisheries Management

Course Title: Seminar; Course code: FMG 598; Term: Winter’22

BANGABANDHU SHEIKH MUJIBUR RAHMAN AGRICULTURAL UNIVERSITY

GAZIPUR-1706

Antibiotics Toxicity in Fish and Its Impact on Public Health[1]

By

Anasua Kar[2]

ABSTRACT

Antibiotics are the common option to minimize microbial disease in super-intensive aquaculture systems. Antibiotics are used for the both therapeutic and prophylactic purpose. But the use of improper doses, unauthorized antibiotics, self-suggested antibiotics and not maintaining accurate withdrawal period after applying antibiotics make the antibiotics toxic for fish as well as human. This review paper summarized antibiotics used in aquaculture, its toxicity to fish and public health. Oxytetracycline, ciprofloxacin and amoxicillin are the most frequently used antibiotics in the aquafarm in Bangladesh, whereas antibiotics from quinolones group are mostly used in global aquaculture. The toxic effects of antibiotics to fish are oxidative stress, hematological and biochemical changes in blood, neurotoxicity as well as genotoxicity and apoptosis. Antibiotic residue in fish constitute antibiotic resistant genes which are very alarming for both fish and human. Due to antibiotic treated fish consumption, people can face health problems such as, allergy, toxicity, and antibiotic resistance. Alternative use of antibiotics such as probiotics was found as a worthy invention for lowering antibiotic use in aquaculture. Careful use of probiotics can protect the aquaculture sector and public health risks might be minimized.

Keywords: Antibiotic toxicity, antibiotic resistance, aquaculture, public health

---

[1]Title of the seminar paper presented as a part of the course, FMG 598 during Winter 2022

[2]MS student, Department of Fisheries Management, BSMRAU, Gazipur-1706

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

With a contribution of 87.5 million tons of fish to global production, aquaculture is one of the most pledging industries among those that produce food (FAO, 2022). According to the FAO, there would be a 4.4 percent drop in fisheries catch in 2020 as a result of lower pelagic species catches and the COVID-19 pandemic's effects (FAO, 2022). Due to aquaculture's ongoing expansion, this declining trend was offset (FAO, 2022). Besides that, the increasing population demands more protein so the increased demand for protein creates pressure on intensive culture methods. As a result, aquaculture in Bangladesh is constantly getting more intensive and commercialized (Shamsuzzaman *et al*., 2017). Compact, overcrowded aquaculture systems allows the rapid spread of diseases. Ultimately fish production is limited by reducing growth and survival rates which heavily compromises the economy (Zhang *et al*., 2021). Hence, in order to prevent production loss and enhance the health status of fish, hatchery owners utilize antibiotics for preventative, therapeutic, and metaphylactic purposes (Cabello *et al*., 2013).

Antibiotics have not always been used responsibly in aquaculture, and control of their use has not provided proper assurance of the prevention of risks to humans (FAO, 2003). Unregulated use of antibiotics in the aquaculture industry for the production of farm-raised fish and shrimps could impose human health and food safety concerns that remain largely unaddressed in most developing nations of the world. The presence of antibiotic residues in aquaculture products could result in the development of bacterial resistance and toxicity to consumers which can lead to morbidity or death (Okocha *et al*., 2018). Incorrectly administered antibiotics - a key factor associated with the development of antimicrobial resistance. Antimicrobial resistance can be disseminated rapidly within microbial ecosystems via mobile genetic elements, posing a risk for humans and animals infected with antimicrobial resistant pathogens as treatments with antibiotics become ineffective (Bell *et al*., 2023). Toxic effects of antibiotics include immunopathological effects, carcinogenicity, mutagenicity, nephropathy, genotoxicity, apoptosis, and neurotoxicity.

However, there were 30 antibiotics recorded in the Narsingdi, Bangladesh area with various trade names and utilized by fish farmers, and most farmers are found ignorant about the mode of action and dose of a particular chemical in the Narsingdi district in Bangladesh (Kawser *et al*., 2022). As a result, there is a chance of antibiotic residue in fish causing antibiotic toxicity and hazards to human health. But the amount of residue concentration, toxicity evaluation, and health risk assessment including antibiotic resistant genes, have not been studied yet. About 14 branded antibiotics with different trade names were seen in the market as well as used by the fish farmers in the southwest coast of Bangladesh for shrimp culture (Shamsuzzaman & Biswas, 2012). But authorization and risk of their use in aquaculture were not identified. Even, antibiotics were seen to be used indiscriminately in the fish farms without knowing the exact reasons of disease (Rahman *et al.*, 2017). However, antibiotic residues in fish also constitute a concern to human health, producing allergic reactions, toxicity, and changes in gut flora, among others which may compromise the immune system in the post-covid-19 era (Edeh & Nsofor., 2023).

Effects are being evaluated in relation to current uses of antibiotics in aquaculture, and if alternative antibiotic use is established, long-term toxic effects of antibiotics on fish and humans may be mitigated.

With these considerations in regard, the present review study is aimed with the following objectives-

i. To review the present condition of antibiotics used in aquaculture
ii. To assess the toxicological effects of antibiotics in fish and the potential public health risk associated with antibiotics treated fish consumption

CHAPTER II

MATERIALS AND METHODS

The main focus of this seminar paper is a review. Therefore, all the data and information presented in this paper are mainly collected from the secondary sources. Secondary sources include a variety of papers, journals, and articles that have been already published. The data was also collected from different relevant reports, and websites which are available on online platforms.

I was able to make this paper better with the help of valuable criticism and suggestions from my major professor, and the course instructors. Afterward, all of the collected information was arranged according to the sequence and presented in this paper.

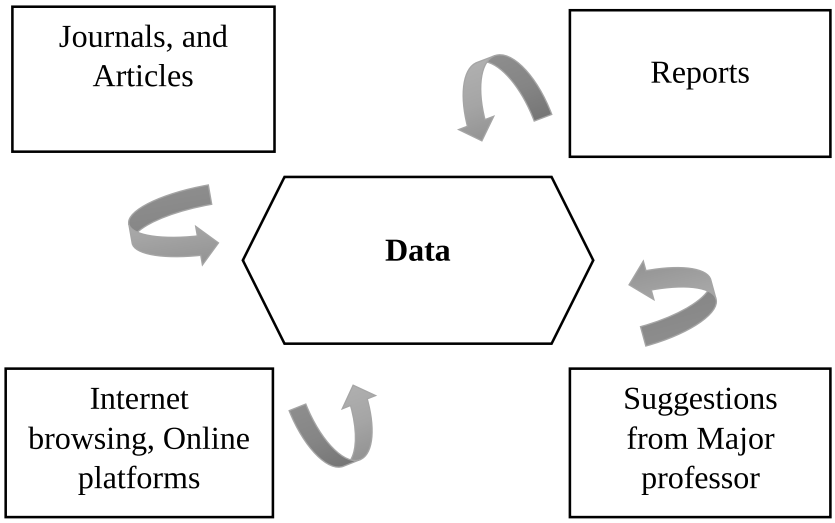

Figure 1. Sources of data and information used to prepare this paper.

CHAPTER III

REVIEW OF FINDINGS

Antibiotic toxicity refers to when antibiotics cause any toxicity such as neurotoxicity, genotoxicity, and developmental disorders in fish. Due to their selective toxicity, antibiotics must not only be very efficient against the bacterium but also have little side effects on both humans and animals (Yang *et al*., 2020). Side effects depend on the doses and user administration methods of antibiotics.

3.1 Present status of antibiotics used in aquaculture

Aquaculture farming methods, various local and governmental regulations, and regulatory enforcement capacity all affect the nature and quantity of antibiotics used. In response to the global concern about antibiotic resistance and the consumption of residues in food, certain nations have already adopted stronger rules. Even though, the greatest of aquaculture production occurs in nations with poor environmental protections and "permissive rules" (Edeh & Nsofor., 2023).

3.1.1 Antibiotics used in major aquaculture producing countries

The major aquaculture producing countries are, China, Indonesia, India, Vietnam, Philippines, Bangladesh, South Korea, Norway, Chile, Egypt, Italy, Japan, Myanmar, Thailand, Brazil, Malaysia, Sri Lanka, Taiwan, UK, and USA (Lulijwa *et al*., 2020; Chen *et al*., 2020). Antibiotics are used to protect fish from disease and to increase fish production. All of the countries are not using authorized antibiotics and also the doses of antibiotics are not as the guideline by the authority.

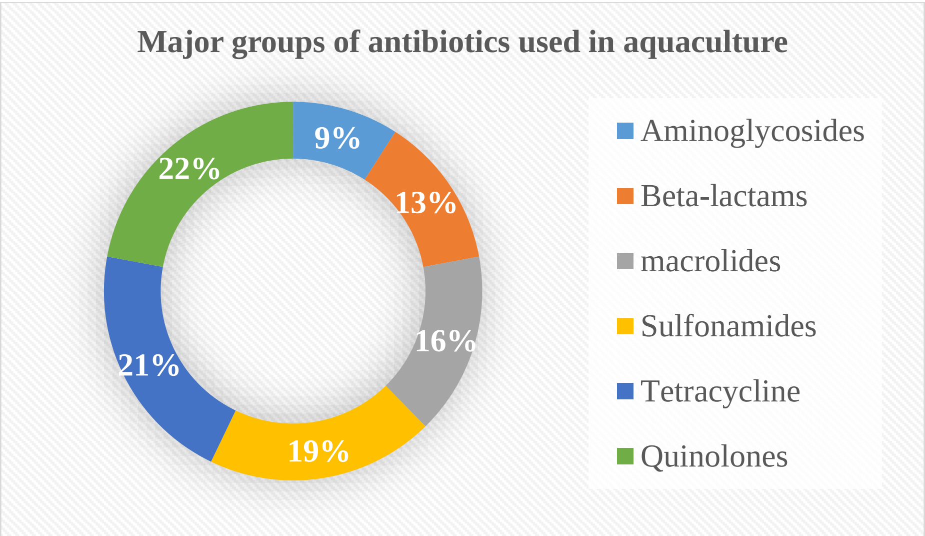

Figure 2. Major groups of antibiotics used in aquaculture (Source: Hossain *et al*., 2022).

The common groups of antibiotics that are used by the major aquaculture producing countries are aminoglycosides, beta-lactams, macrolides, sulfonamides, tetracycline, quinolones, etc. It is found that quinolones are the most commonly used antibiotics group in which ciprofloxacin, enrofloxacin, oxolinic acid, norfloxacin, and flumequin are the mostly used antibiotics in different countries (Hossain *et al*., 2022).

3.1.2 Authorized antibiotics for aquaculture uses

All antibiotics are not suitable for aquatic organisms. Only approved antibiotics should be applied to fish and other aquatic organisms. Therefore, authorities have approved some antibiotics with the dose for use in aquaculture.

Table 1. List of authorized antibiotics for aquaculture use in major aquaculture producing countries

Country name	Authorized antibiotics		References
	Number of antibiotics	Antibiotics name	
China	13	DC, ENF, FLF, FMQ, NEO, NRF, OLA, SDZ, SMZ, SMX, SMM, THP, TMP	(Liu *et al*., 2017; Lulijwa *et al*., 2020)
Chile	19	AMX, CHP, DC, ENF, ERY, FLF, FMQ, FZD, CN, NEO, NRF, OLA, OTC, SDZ, SMZ, SMX, SMM, THP, TMP	(Preena *et al*., 2020)
Vietnam	30	AMX, BZP, CIP, CLX, CT, CTC, CYM, DNF, DLX, DFX, EMM, ERY, FMQ, NEO, OLA, OTP, OTC, OX, PRM, SAF, SDMX, SDZ, SMM, SMX, SMZ, SPM, TC, TLC, TMP, TYL	(Lulijwa *et al*., 2020; Mard, 2014)
South Korea	17	AMX, CIP, CTC, ENF, ERY, FLF, FMQ, NDA, OTP, OLA, OTC, SDZ, SCP, SMX, SDMX, THP, TMP	(Lulijwa *et al*., 2020)
Thailand	14	AMX, ENF, NRF, OTC, OTP, PEN, SDZ, SDMX, SMM, SDMX+, SGD, TMP, TBS, TC	(Lulijwa *et al*., 2020)
Bangladesh	12	AMX, CTC, DC, ERY, OTC, PEN-G, SDZ, SMZL, SMX, SMZ, THP, TYL	(Hossain *et al*., 2017; Ali *et al*., 2016)
Japan	11	AMX, CA, DC, ERY, FFM, LIN, OLA, OTC, SMM, SAS, THP	(Chen *et al*., 2020)
Italy	6	AMX, FMQ, OTC, TEC, TMP, SDZ	(Preena *et al*., 2020)
United Kingdom	5	AMX, Co-TMZ, FLF, OTC, OLA, SAF	(Preena *et al*., 2020)
America	4	FLF, OTC, TMP, SMP, SDMX	(Preena *et al*., 2020)

*AMX= Amoxicillin; BZP= Benzylpencillin; CA= Carbolic acid; CHP= Chloramphenicol; CIP=Ciprofloxacin; CLX= Cloxacillin; CN= Gentamycin; CT= Colistin; CTC= Chlortetracycline; CYM= Cypermethrim; Co-TMZ= Co-trimazine; DC= Doxyxycline; DFX= Difloxacin; DLX= Dicloxacillin; DNF= Danofloxacin; EMM= Emamecyin; ENF= Enrofloxacin; ERY= Erythromycin; FFM= Fosfomycin; FLF= Florfenicol; FMQ= Flumequin; FZD= Furozolidin; LIN= Lincosamide; NDA= Nalidixic acid; NEO= Neomycin; NRF= Norfloxacin; OLA= Oxolinic acid; OTC= Oxytetracycline; OTP= Ormethoprim; OX= Oxacillin; PEN= Penicillin; PRM= Paromomycin; SAF= Sarafloxacin; SAS= Sodium alkane sulfonate; SCP= Sulfachloropyridazine; SDMX= Sulfadimethoxine; SDZ= Sulfadiazine; SGD= Sulfaguanidine; SMM= Sulfamonomethoxine; SMP= Sulfatrimethoprim; SMX= Sulfamethoxazole; SMZ= Sulfamethazine; SMZL= Sulfamethizole; SPM= Spectinomycin; TBS= Tribrissen; TC= Tetracyclin; THP= Thiamphenicol; TLC= Tilmicosin; TMP= Trimethoprim; TYL= Tylosin

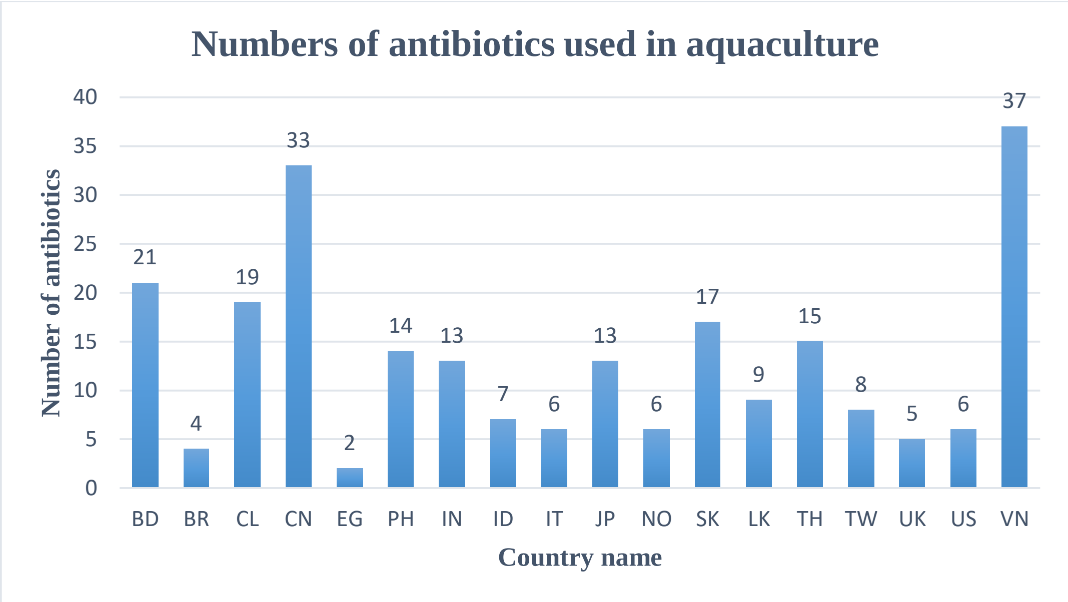

Figure 3. Numbers of antibiotics used in major aquaculture-producing countries. BD= Bangladesh; BR= Brazil; CL= Chile; CN= China; EG= Egypt; PH= Philippines; IN= India; ID= Indonesia; IT= Italy; JP= Japan; NO= Norway; SK= South Korea; LK= Sri Lanka; TH= Thailand; TW= Taiwan; UK= United Kingdom; US= United States of America; VN= Vietnam (Source: Hossain *et. al*., 2022).

3.1.3 Currently used antibiotics in Bangladesh

To maintain fish health in intensive aquaculture, fish farmers are using antibiotics frequently in their fish farms. Among them, aminoglycosides, beta-lactams, macrolides, sulfonamides, tetracycline, quinolones, and some other groups are included (Table 2). Most commonly used antibiotics are from the sulfonamides group. Although many of these antibiotics are banned, farmers still use them to control fish diseases. Chowdhury *et al*., (2022) found that oxytetracycline are used frequently than the other antibiotics in aquafarms.

Table 2. Antibiotics used in Bangladesh

Major groups of antibiotics	Antibiotics name	References
Aminoglycosides	Neomycin, Kanamycin	(Chowdhury *et al*., 2022; Ali *et al*., 2016; Hossain *et al*., 2017; Hossain *et al*., 2018; Kawsar *et al*., 2022)
Beta-lactams	Amoxicillin, Penicillin	
Macrolides	Erythromycin, Tylosin	
Sulfonamides	Sulfadiazine, Sulfamethoxazole, Sulfamethazine, Trimethoprim, Tribrissen, Cortimoxazole, Sulfamethizole	
Tetracycline	Chlortetracycline, Oxytetracycline, Doxycycline	
Quinolones	Ciprofloxacin, Enrofloxacin, Levofloxacin	
Others	Choramphenicol, Furazolidon, Furadin, Nitrofurantoin	

More specifically, mostly used antibiotics in Bangladesh are oxytetracycline, amoxicillin, and ciprofloxacin in farm-level aquaculture (Figure 04).

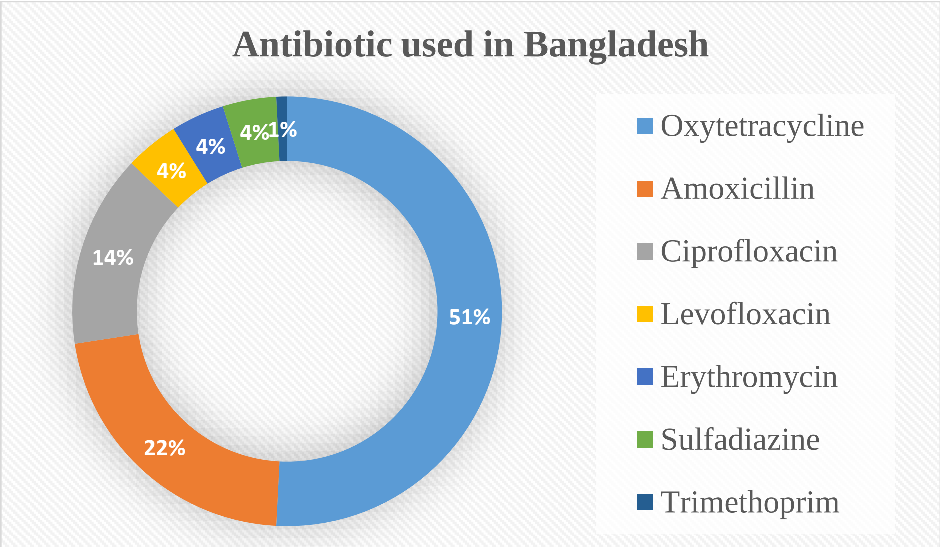

Figure 4. Farm-level antibiotics used in Bangladesh (Source: Chowdhury *et al*., 2022).

3.2 Antibiotics accumulation in fish and human

3.2.1 Possible sources of antibiotics in aquatic environment and accumulation in fish

Overuse of antibiotics and disposal of their residues from various sources have increased throughout the past 20 years despite the lack of legislation surrounding these issues (Sosa-Hernandez *et. al*., 2021). Antibiotics that can be detrimental to fish reach the aquatic environment through a variety of sources, such as manufacturing waste and human and veterinary applications (Yang *et al*., 2020).

When fishes are exposed to an aquatic environment containing antibiotics, the antibiotics can enter the fish's body. Moreover, when antibiotics are given to fish through food, the fish may accumulate antibiotics in their body due to overdoses. Antibiotics can accumulate in fish by bath treatment for not maintaining withdrawal period. Vulnerable organs of fish in which antibiotics may accumulate are, liver, intestine as well as fish muscle (He *et al*., 2014).

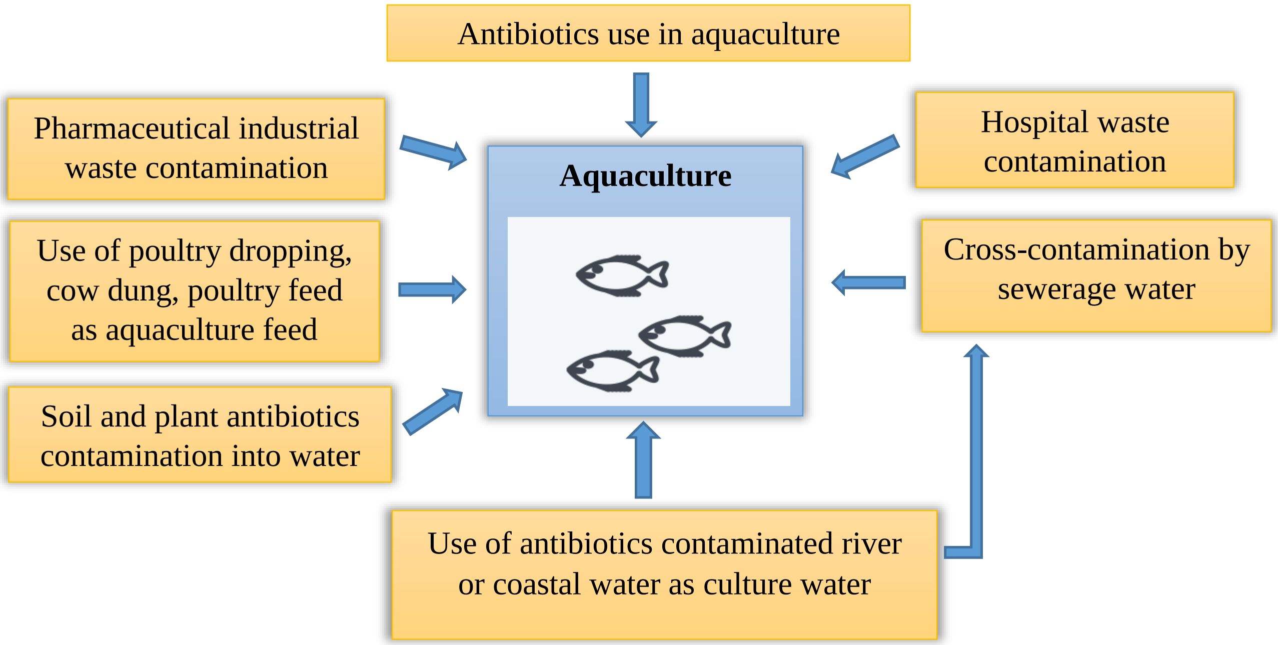

Figure 5. Schematic diagram of probable sources of antibiotic contamination in aquaculture (Source: Hossain *et al*., 2022; Sosa-Hernandez *et al*., 2021).

3.2.2 Antibiotics exposure pathway in human through aquaculture

Fish handling and consumption are the direct pathways of human exposure to antibiotics from fish. And antibiotic resistance can also be introduced to human body pathogens via fish consumption. Antibiotics can enter humans through contaminated water, consumption of antibiotic treated fish, handling of fish during antibiotic treatment and the food chain (Kawsar *et al*., 2022). As a result, there is a possibility of public health risk. Because unnecessary induction of antibiotics in human body can cause side effects on public health.

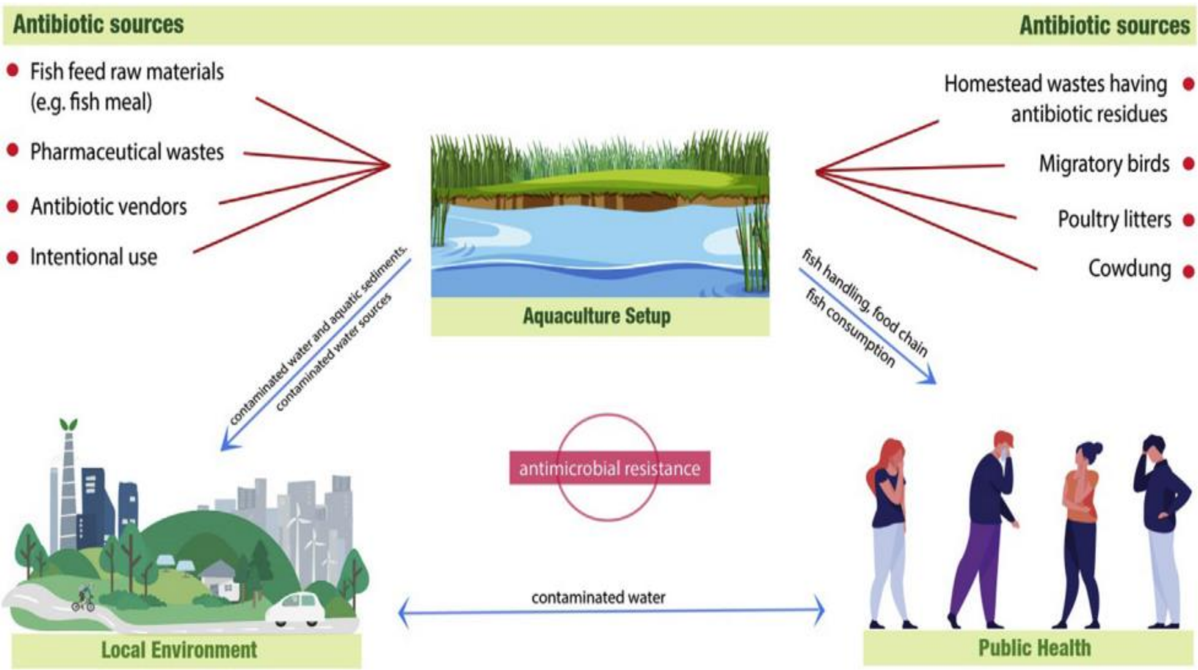

Figure 6. The possible exposure pathway of antibiotics in humans through aquaculture (Source: Kawsar *et al*., 2022).

3.3 Toxicological effects of antibiotics in fish

3.3.1 Antibiotics causes toxicity in fish

All antibiotics are not toxic for each species. Such as, ciprofloxacin, ofloxacin, norfloxacin, enrofloxacin, sulfamethazine, clarithromycin, and tilmicosin found toxic at specific doses in case of *Danio rerio* (Wang *et al*., 2014; Yin *et al*., 2014; Yan *et al*., 2018; Ryan *et al*., 2017; Yan *et al*., 2019). Toxic effects include abnormal development, histopathological changes, abnormal heart rate, abnormal hatching, metabolic changes, oxidative stress, and apoptosis. Some other antibiotics that causes toxicity are, sulfamethoxazole, oxytetracycline, erythromycin, roxythromycin. Their toxic effects include alteration of nutrient metabolism, innate immune system, altered activity of anti-oxidant enzymes, and neurotoxicity. Some literature reported about the toxic effects on certain species by specific antibiotics doses (Table 3).

Table 3. List of reported antibiotics that cause toxicity in fish

Group of antibiotics	**Name of antibiotics**	**Concentrations (weight/volume)**	**Species**	**Toxicity**	**Reference**
Quino-lones	Ciprofloxacin, ofloxacin, norfloxacin and Enrofloxacin	>37.5 mg/L 4.69, 9.38 mg/L	*Danio rerio*	Abnormal development and histo- pathological changes	(Wang *et al.*, 2014; Yin *et al.*, 2014)
Sulfona-mides	Sulfa-methazine	0.2-2000 μg/L	*Danio rerio*	Abnormal heart rate and hatching	(Yan *et. al.*, 2018)
	Sulfa-methoxazole	260 ng/L	*Oreochromis niloticus*	Alteration of nutrient metabolism and innate immune system	(Limbu *et. al.*, 2018)
Tetra-cyclines	Oxytetra-cycline	100 mg/kg	*Oncorhynchus mykiss*	Oxidative stress	(Zargar *et al.*, 2020)
Macrolides	Erythromycin	2 μg/L	*Carassius auratus*	Altered activity of anti-oxidant enzyme	(Rodrigues *et al.*, 2016)
	Roxithro-mycin	50 μg/L	*Oreochromis niloticus*	Neurotoxicity	(Zhang *et. al.*, 2019)
	Clarithro-mycin	0.1 mg/L	*Danio rerio*	Metabolic changes	(Ryan *et. al.*, 2017)
	Tilmicosin	40 mg/L	*Danio rerio*	Oxidative stress and apoptosis	(Yan *et al.*, 2019)

Oxytetracyclin, florfenicol and trimethoprim are found more frequently in fish muscle as residue. Besides that, sulfamethoxazole, sulfamethazine, enrofloxacin, ciprofloxacin also presents in fish muscle as residue which causes toxicity to fish.

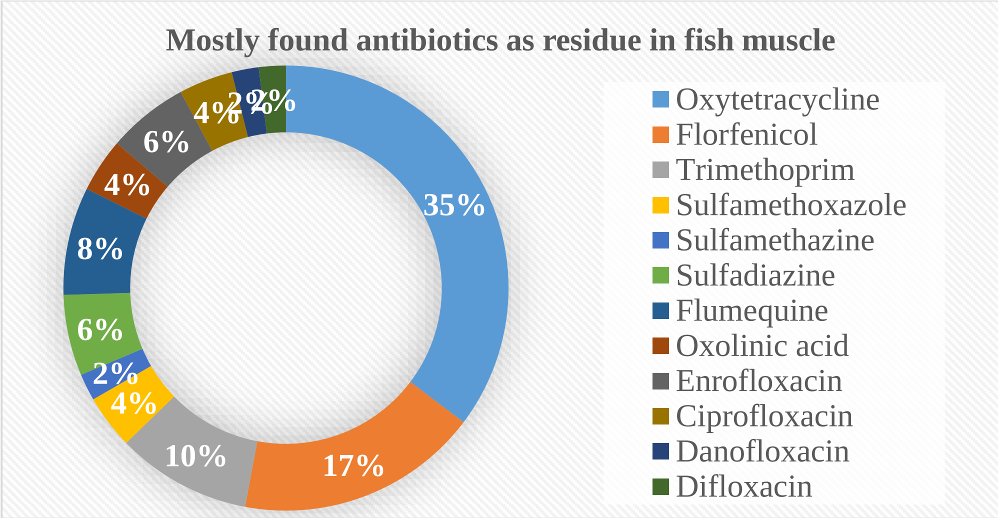

Figure 7. Antibiotic residue (%) found in various fish muscle (Source: Limbu *et al.*, 2021).

3.3.2 Toxicological effects in fish

3.3.2.1 Oxidative stress

Oxidative stress mainly refers to the imbalance between reactive oxygen species (ROS) production and antioxidant defense systems in organisms. Excessive oxidative stress could induce cell and tissue damage, mainly manifested as DNA hydroxylation, protein denaturation, lipid peroxidation and cell apoptosis (Hoseinifar *et al*., 2020). Antibiotic use in aquaculture stimulates excessive endogenous ROS production, which leads to oxidative stress. Malondialdehyde (MDA) is a key indicator of lipid peroxidation since it is the primary oxidative product of peroxidized polyunsaturated fatty acids (Limbu *et al*., 2021). Thiobarbituric acid reactive substances, which are indicated as MDA concentrations (Nunes *et al*., 2015). The increased lipid peroxidation may result in hemolysis, the breakdown of tissues, harm to the nervous system, a general decline in cellular metabolism, and ultimately cell death (Liu *et al*., 2015).

Table 4. Effects of antibiotics on lipid peroxidation and oxidative stress in cultured fish

Antibiotic	Dose	Parameter	Fish species	Effect	Reference
Oxytetracycline	82.5 mg/kg B.W/day	MDA intestine	*Oreochromis niloticus*	↑	(Limbu *et al*., 2019)
Sulfamethoxazole	100 mg/kg B.W/day	MDA liver	*Oreochromis niloticus*	↑	(Limbu *et al*., 2018)
Norfloxacin	0.0001, 0.1, 5, 10 mg/L	TBARS	*Cyprinus carpio*	↓	(Charvatova *et al*., 2015)
Enrofloxacin	50, 100, 200 mg/ml	MDA	*Ctenopharyngodon idellus*	↑	(Liu *et al*., 2015)
Tetracycline	5, 50, 500 ng/L	TBARS	*Gambusia holbrooki*	↓	(Nunes *et al*., 2015)
Ciprofloxacin	0.7, 1100, 3000 µg/L	TBARS	*Danio rerio*	↓	(Plhalova *et al*., 2014)
Sulfamonomethoxine	0.01, 1, 100 µg/L	ROS	*Danio rerio*	↑	(Qiu *et al*., 2020)

The orange arrow shows decreased parameters in treated fish compared to control, the blue arrow shows higher values in treated fish. Malondialdehyde (MDA), thiobarbituric acid reactive substances (TBARS), superoxide dismutase (SOD), glutathione-S-transferase (GST), reduced glutathione (GSH) and reactive oxygen species (ROS), Body weight (B.W)

3.3.2.2 Hematological and blood biochemical changes

The health of farmed fish after antibiotic exposure can be determined in large part by hematological markers. The combined effects of feeding and bath antibiotic exposure on farmed fish impaired hematological indicators, suggesting long-term toxic consequences that damage the fish's health and aerobic metabolism (Limbu *et al*., 2021). However, some literature demonstrates the effects of antibiotics on hematological parameters (Table 5).

Antibiotics can change hematological parameters in cultured fish. Oxytetracyclin reduce red blood cell count, increase white blood cell count in *Labeo rohita* at 80 mg/L dose (Ambili *et al*., 2013). Oxytetracyclin also reduce lysozyme, neutrophils and white blood cell count in *Oncorhynchus mykiss* (Hoseini and Yousefi, 2019). Chloramphenicol and metronidazole changes lymphocytes, total white blood cell count, lysozyme activity (Nwani *et al*., 2014; Han *et al*., 2014).

Table 5. Effects of antibiotics on hematological parameters in cultured fish

Antibiotic	Dose	Parameter	Fish species	Effect	Reference
Oxytetracycline	80 mg/L	RBC count	*Labeo rohita*	↓	(Ambili *et al*., 2013)
Oxytetracycline	80 mg/L	WBC	*Labeo rohita*	↑	(Ambili *et al*., 2013)
Oxytetracycline	2.5 g/kg b.w/day	Lysozyme, Neutrophils, WBC	*Oncorhynchus mykiss*	↓	(Hoseini and Yousefi, 2019)
Chloramphenicol	2.5, 5 and 10 mg/L	Lymphocytes	*Clarias gariepinus*	↑	(Nwani *et al*., 2014)
Metronidazole	0.1, 0.5, or 2.5 mg/L	Total WBC count, complement activity, lysozyme activity and bactericidal activity	*Cyprinus carpio*	↓	(Han *et al*., 2014)

Orange arrow indicates decreased parameters and blue arrow indicates increased parameters in treated fish compared to control. RBC= red blood cell; WBC= white blood cell

3.3.2.3 Neurotoxicity

In general, antibiotics exposure in cultured fish cause neurotoxicity depending on dose, time and exposure methods. Because of their increased bioavailability, simpler digestion, and quicker absorption rates, bath exposure results in greater neurotoxicity than food remedies (Limbu *et al*.,2018). Antibiotics causes Reduced Acetylcholinesterase enzyme(AChE) activity which Inhibit Ca^{+} ion at a common receptor site on the nerve ending membrane. As a result, neuronal and muscle injury occur and nervous system is interrupted. Ultimately antibiotic causes neurotoxicity in fish (Rhee *et al*., 2013; Liu *et al*., 2014; Chow *et al*., 2004).

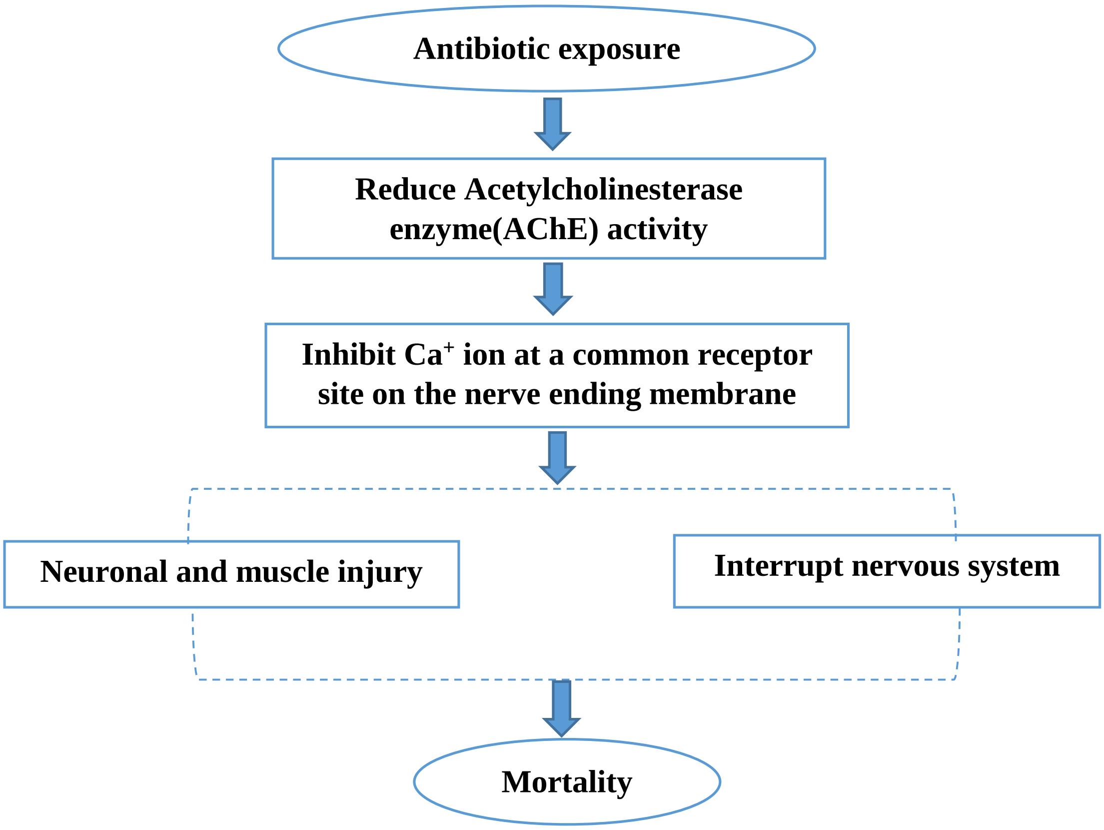

Figure 8. A schematic diagram of neurotoxicity of fish due to antibiotics (Source: Rhee *et al*., 2013; Liu *et al*., 2014; Chow *et al*., 2004).

3.3.2.4 Genotoxicity and apoptosis

The term "genotoxicity" describes a new chemical compound's capacity to change genetic information (Vasdev *et al*., 2022). Antibiotics are capable of generating reactive oxygen species (ROS) (Rodrigues *et al*., 2016), oxidation continues in the presence of transition metal ions, high DNA binding affinity alters the secondary structure of Natural DNA, causing genotoxicity in fish (Rodrigues *et al*., 2017).

Apoptosis, a programmed cell death in which cell destroy themselves can be induced due to higher concentration of antibiotic in cell. Such as, higher concentrations of doxycyclines can induce apoptosis in cell (Shan *et al*., 2022). Sulfamethoxazole raised the activity of apoptosis-related genes in a dose-dependent manner (Iftikhar *et al*., 2023). Besides that, exposing *Danio rerio* to toxicological levels of tetracyclines increased ROS production, which increased apoptosis (Zhang *et al*., 2015). These studies show that the immune system disruption and excessive ROS production caused by antibiotics in different fish species can result in apoptosis (Limbu *et al*., 2021).

In general, antibiotics used on cultured fish result in chromosomal anomalies and/or spillage, which trigger genotoxicity and apoptosis. These effects are harmful because they directly affect genetic materials, affecting fish integrity and leading to mutations with terrible effects for overall aquaculture production (Limbu *et al*., 2021).

3.4 Potential health risk in human through fish consumption

Concern over public health has increased due to the consumption of antibiotic-treated fish worldwide (Limbu *et al*., 2021). Antibiotics generally pose a risk to human health in two ways: adverse drug reactions (ADRs) and the potential spread of antibiotic resistance by exerting selective pressure on clinically significant bacteria (Liu *et al*., 2017). The duration of exposure determines the risk that antibiotics pose to human health (Limbu *et al*., 2021). It is necessary to evaluate the health risk to consumers due to antibiotic residues in commercial fish and the presence of some antibiotics that exceed maximum residue limits (Shaaban & Mostafa, 2023). Evaluations regarding direct risks to human health from consuming antibiotic treated fish are limited.

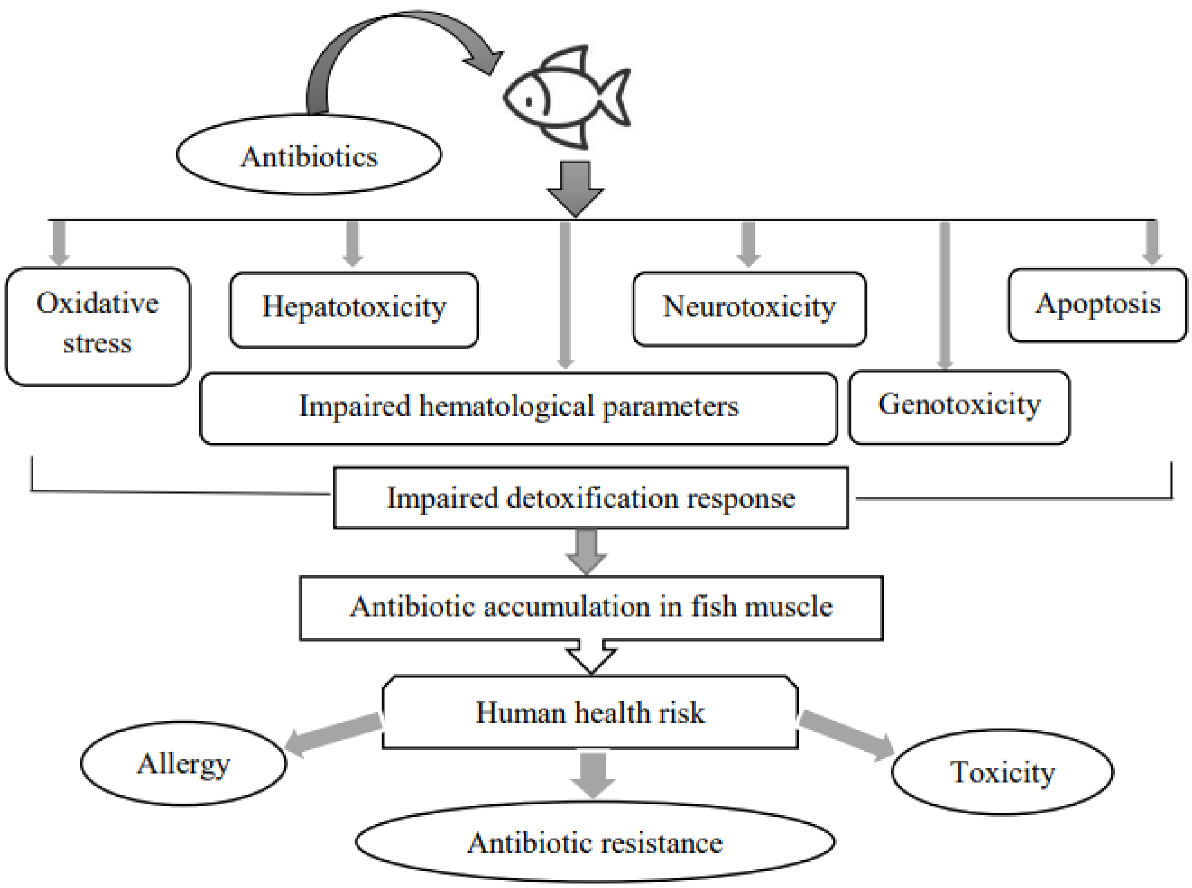

Figure 9. A systemic effects of antibiotics in cultured fish and public health through antibiotic treated fish consumption (Source: Limbu *et al.*, 2021).

3.4.1 Antibiotic resistance

Antibiotics also pose a risk to the public's health because they can spread from aquatic organisms to people through the development of antibiotic-resistant bacteria (ARB) and genes (ARGs) (Chen *et al.*, 2020). A high diversity of ARGs found in finfish ponds in the Mymensingh division of Bangladesh, with the most common being aminoglycosides and sulfonamides resistance genes (Bell *et al.*, 2023).

Table 6. Antibiotic resistant bacteria and resistant antibiotics associated with human pathogens

Resistant bacteria	Resistant antibiotic group	Resistant antibiotic	Reference
Pseudomonas spp.	Floroquinolone	-	(Bell *et al.*, 2023)
Klebsiella pneumonia, *Escherichia coli*, *Salmonella enterica*	Sufonamide	-	(Bell *et al.*, 2023)
Pseudomonas aeruginosa	Carbapenem, penem, cephalosporin, cephamycin	-	(Bell *et al.*, 2023)
Mycobacterium spp.	Fosfomycin	-	(Bell *et al.*, 2023)
Vibrio parahaemolyticus		Ampicillin, chloramphenicol, erythromycin, gentamicin, nitrofurantoin, oxolinic acid, tetracycline, ciprofoxacin, polymyxin- B	(Haque *et al.*, 2015)
Aeromonas spp.		Ampicillin, erythromycin, cephalothin, nalidixic acid, streptomycin, tetracycline, chloramphenicol, and gentamicin	(Rahman *et al.*, 2009)

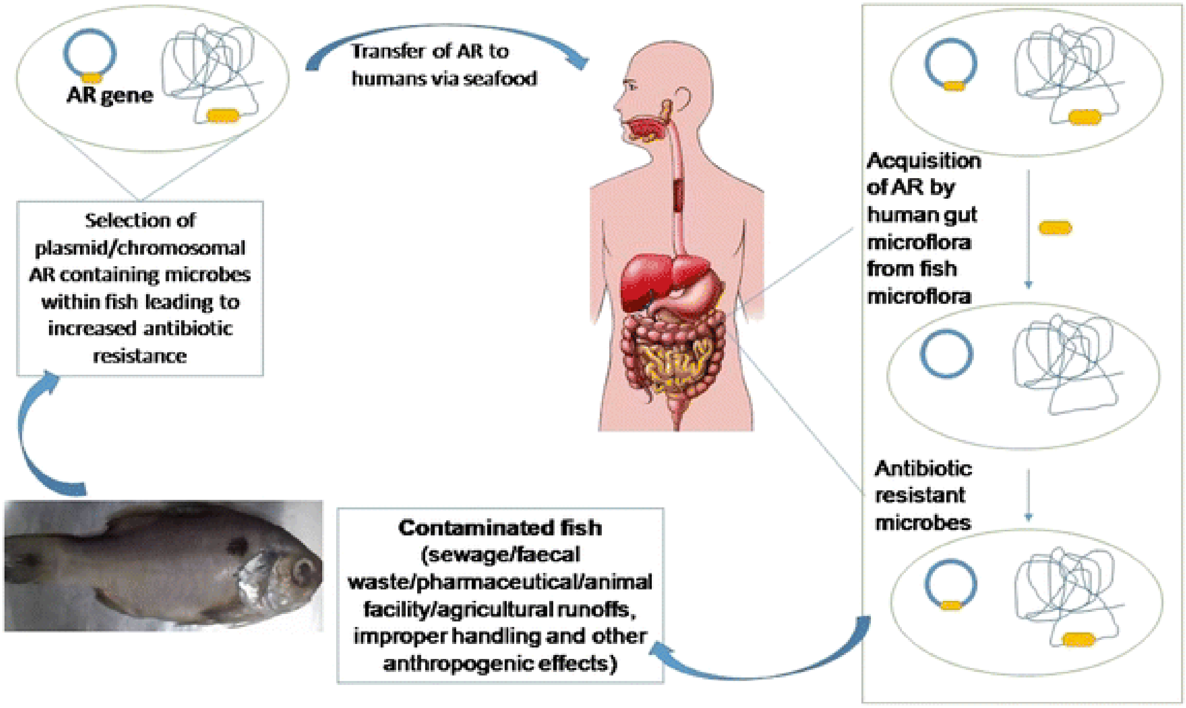

Figure 10. Spread of antibiotic-resistant traits (ART) from fish to humans (Source: Naik *et al*., 2018).

3.4.2 Allergy

Generally, occurrence of antibiotics residues in edible fish may be responsible for allergic reactions, bacterial resistance and toxic effects on human health (Shaaban & Mostafa, 2023). The higher seafood consumption per kilogram of body weight was mainly responsible for the higher antibiotics estimated daily intake (EDIs) exposure to children (Wang *et al*., 2023). Most antibiotics (e.g., Tetracyclines, Salphonamides) are antigenic (Li, 2008). If residues of these antibiotics remain in the edible fish muscle and thereby consumed by human than allergic reaction can be occurred.

3.4.3 Miscellaneous toxicity

Impact of toxicity include carcinogenicity, mutagenicity, teratogenicity, bone marrow suppression, and destruction of normal intestinal flora (Liu *et al*., 2017; Okocha *et al*., 2018). Maternal use and child's use of cephalosporins, sulphonamides and trimethoprim, macrolides and amoxicillin may increase risk of asthma in children, and the class of cephalosporins has the strongest association with asthma (Metsala *et al*., 2015). Sulfonamodes that enter human bodies through consumption would destroy the human hematopoietic system, and consequentially cause hemolytic anemia (He *et al*., 2014).

CHAPTER IV

CONCLUSIONS AND RECOMMENDATIONS

This review paper represents the overall antibiotics used in aquaculture as well as the toxicity of antibiotics with the impact of public health.

The occurrence and accumulation of antibiotics residues in the aquatic organisms as well as aquatic environment is nowadays a burning issue. It is not avoidable because it is related to fish health and most importantly human health. Antimicrobials are the easy option for treatment of microbial diseases in fish. Antibiotics from quinolones group are mostly used in aquaculture. In Bangladesh oxytetracycline, ciprofloxacin and amoxicillin are the most frequently used antibiotics in aqua farm. But all of them are not authorized for aquaculture use as well as their improper and heedless use can be harmful and toxic for fish.

The toxic effects are oxidative stress, hematological and blood biochemical changes, neurotoxicity, genotoxicity and apoptosis. Besides that, the residues that remains in the fish body can grow antibiotic resistance strains of bacteria that can be a threat for human health. Because these antibiotic resistant genes are spreading into human through consumption of antibiotic treated fish. In Bangladesh aminoglycosides and sulfonamides resistance genes are isolated more frequently in fish which can raise antibiotic resistance in human body who consume these fish. Human can face allergic reactions, toxicity, and antibiotic resistance due to consuming antibiotic treated fish.

Aquaculture operators should adopt alternatives to antibiotics in treating and preventing fish disease. As the alternative use of antibiotics probiotic use is recommended in many research work. That is a good and harmless way to improve immune system of aquatic organisms which can avoid frequent antibiotic use in aquaculture for disease treatment. Further studies are needed, to assess the extent to which antibiotic use is directly correlated with human health problems and reduced antibiotic effectiveness in fish farms in Bangladesh.

CHAPTER V

REFERENCES

Ali, H., Rico, A., Murshed-e-Jahan, K., & Belton, B. (2016). An assessment of chemical and biological product use in aquaculture in Bangladesh. *Aquaculture, 454*, 199-209.

Ambili, T. R., Saravanan, M., Ramesh, M., Abhijith, D. B., & Poopal, R. K. (2013). Toxicological effects of the antibiotic oxytetracycline to an Indian major carp *Labeo rohita*. *Archives of Environmental Contamination and Toxicology*, *64*, 494-503.

Bell, A. G., Thornber, K., Chaput, D. L., Hasan, N. A., Alam, M. M., Haque, M. M., & Tyler, C. R. (2023). Metagenomic assessment of the diversity and ubiquity of antimicrobial resistance genes in Bangladeshi aquaculture ponds. *Aquaculture Reports*, *29*, 101462.

Cabello, F. C., Godfrey, H. P., Tomova, A., Ivanova, L., Dolz, H., Millanao, A., & Buschmann, A. H. (2013). Antimicrobial use in aquaculture re-examined: its relevance to antimicrobial resistance and to animal and human health. *Environmental Microbiology*, *15*(7), 1917-1942.

Charvatova, N., Zelinska, G., Dobsikova, R., Stancova, V., Zivna, D., Plhalova, L., & Svobodova, Z. (2015). The effect of the fluoroquinolone norfloxacin on somatic indices and oxidative stress parameters in early stages of common carp (*Cyprinus carpio* L.). *Neuroendocrinology Letters*, *36*(Suppl 1), 79-87.

Chen, J., Sun, R., Pan, C., Sun, Y., Mai, B., & Li, Q. X. (2020). Antibiotics and food safety in aquaculture. *Journal of Agricultural and Food Chemistry*, *68*(43), 11908-11919.

Chow, K. M., Szeto, C. C., Hui, A. C. F., & Li, P. K. T. (2004). Mechanisms of antibiotic neurotoxicity in renal failure. *International Journal of Antimicrobial Agents*, *23*(3), 213-217.

Chowdhury, S., Rheman, S., Debnath, N., Delamare-Deboutteville, J., Akhtar, Z., Ghosh, S., & Chowdhury, F. (2022). Antibiotics usage practices in aquaculture in Bangladesh and their associated factors. *One Health*, *15*, 100445.

Edeh, I. C., & Nsofor, C. I. (2023). Utilization of antibiotics in aquaculture; present status and future alternatives in the post covid-19 pandemic era. *The Bioscientist Journal*, *11*(1), 57-70.

FAO, (2022). The State of World Fisheries and Aquaculture 2022. Towards Blue Transformation. Rome, FAO.

FAO/WHO. Code of Practice for Fish and Fishery Products. Codex Alimentarius Commission. FAO, Rome: CAC/ RCP (2003). pp. 238.

Han, J., Zhang, L., Yang, S., Wang, J., & Tan, D. (2014). Detrimental effects of metronidazole on selected innate immunological indicators in common carp (*Cyprinus carpio* L.). *Bulletin of Environmental Contamination and Toxicology*, *92*, 196-201.

Haque, M. M., Hossain A., Mandal S., Rahman M. S., & Mahmud Z. (2015). Prevalence, characterization and antibiotic susceptibility of *Vibrio parahaemolyticus* isolated from fshes and shellfshes of coastal regions of Bangladesh. *Dhaka Univ J Biol Sci*, 24, 121–129

He, X. T., Wang, Q., Nie, X. P., Yang, Y. T., & Cheng, Z. (2014). Residues and health risk assessment of sulfonamides in sediment and fish from typical marine aquaculture regions of Guangdong Province, China. *Huan Jing ke Xue= Huanjing Kexue*, *35*(7), 2728-2735.

Hoseini, S. M., & Yousefi, M. (2019). Beneficial effects of thyme (*Thymus vulgaris*) extract on oxytetracycline-induced stress response, immunosuppression, oxidative stress and enzymatic changes in rainbow trout (*Oncorhynchus mykiss*). *Aquaculture Nutrition*, *25*(2), 298-309.

Hoseinifar, S. H., Yousefi, S., Van Doan, H., Ashouri, G., Gioacchini, G., Maradonna, F., & Carnevali, O. (2020). Oxidative stress and antioxidant defense in fish: the implications of probiotic, prebiotic, and synbiotics. *Reviews in Fisheries Science & Aquaculture*, *29*(2), 198-217.

Hossain, A., Habibullah-Al-Mamun, M., Nagano, I., Masunaga, S., Kitazawa, D., & Matsuda, H. (2022). Antibiotics, antibiotic-resistant bacteria, and resistance genes in aquaculture: Risks, current concern, and future thinking. *Environmental Science and Pollution Research*, 1-22.

Hossain, A., Nakamichi, S., Habibullah-Al-Mamun, M., Tani, K., Masunaga, S., & Matsuda, H. (2018). Occurrence and ecological risk of pharmaceuticals in river surface water of Bangladesh. *Environmental Research, 165*, 258-266.

Hossain, A., Nakamichi, S., Habibullah-Al-Mamun, M., Tani, K., Masunaga, S., & Matsuda, H. (2017). Occurrence, distribution, ecological and resistance risks of antibiotics in surface water of finfish and shellfish aquaculture in Bangladesh. *Chemosphere*, *188*, 329-336.

Iftikhar, N., Konig, I., English, C., Ivantsova, E., Souders, C. L., Hashmi, I., & Martyniuk, C. J. (2023). Sulfamethoxazole (SMX) Alters Immune and Apoptotic Endpoints in Developing Zebrafish (*Danio rerio*). *Toxics*, *11*(2), 178.

Kawsar, M. A., Alam, M. T., Pandit, D., Rahman, M. M., Mia, M., Talukdar, A., & Sumon, T. A. (2022). Status of disease prevalence, drugs and antibiotics usage in pond-based aquaculture at Narsingdi district, Bangladesh: A major public health concern and strategic appraisal for mitigation. *Heliyon*, *8*(3), e09060.

Li, Z. J. (2008). Advantages and disadvantages and strategies of antibiotic application in aquaculture. *Guizhou Anim. Sci. Vet. Med*, *32*, 23-24.

Limbu, S. M., Chen, L. Q., Zhang, M. L., & Du, Z. Y. (2021). A global analysis on the systemic effects of antibiotics in cultured fish and their potential human health risk: a review. *Reviews in Aquaculture*, *13*(2), 1015-1059.

Limbu, S. M., Ma, Q., Zhang, M. L., & Du, Z. Y. (2019). High fat diet worsens the adverse effects of antibiotic on intestinal health in juvenile Nile tilapia (*Oreochromis niloticus*). *Science of the Total Environment*, *680*, 169-180.

Limbu, S. M., Zhou, L., Sun, S. X., Zhang, M. L., & Du, Z. Y. (2018). Chronic exposure to low environmental concentrations and legal aquaculture doses of antibiotics cause systemic adverse effects in Nile tilapia and provoke differential human health risk. *Environment international*, *115*, 205-219.

Liu, B., Cui, Y., Brown, P. B., Ge, X., Xie, J., & Xu, P. (2015). Cytotoxic effects and apoptosis induction of enrofloxacin in hepatic cell line of grass carp (*Ctenopharyngodon idellus*). *Fish & Shellfish Immunology*, *47*(2), 639-644.

Liu, J., Lu, G., Cai, Y., Wu, D., Yan, Z., & Wang, Y. (2017). Modulation of erythromycin-induced biochemical responses in crucian carp by ketoconazole. *Environmental Science and Pollution Research*, *24*, 5285-5292.

Liu, J., Lu, G., Ding, J., Zhang, Z., & Wang, Y. (2014). Tissue distribution, bioconcentration, metabolism, and effects of erythromycin in crucian carp (*Carassius auratus*). *Science of the Total Environment*, *490*, 914-920.

Lulijwa, R., Rupia, E. J., & Alfaro, A. C. (2020). Antibiotic use in aquaculture, policies and regulation, health and environmental risks: a review of the top 15 major producers. *Reviews in Aquaculture*, *12*(2), 640-663.

Mard, M. (2014). List of drugs, chemicals and antibiotics of banned or limited use for aquaculture and veterinary purposes.

Metsala, J., Lundqvist, A., Virta, L. J., Kaila, M., Gissler, M., & Virtanen, S. M. (2015). Prenatal and post-natal exposure to antibiotics and risk of asthma in childhood. *Clinical & Experimental Allergy*, *45*(1), 137-145.

Naik, O. A., Shashidhar, R., Rath, D., Bandekar, J. R., & Rath, A. (2018). Characterization of multiple antibiotic resistance of culturable microorganisms and metagenomic analysis of total microbial diversity of marine fish sold in retail shops in Mumbai, India. *Environmental Science and Pollution Research*, *25*, 6228-6239.

Nunes, B., Antunes, S. C., Gomes, R., Campos, J. C., Braga, M. R., Ramos, A. S., & Correia, A. T. (2015). Acute effects of tetracycline exposure in the freshwater fish *Gambusia holbrooki*: antioxidant effects, neurotoxicity and histological alterations. *Archives of Environmental Contamination and Toxicology, 68,* 371-381.

Nwani, C. D., Mkpadobi, B. N., Onyishi, G., Echi, P. C., Chukwuka, C. O., Oluah, S. N., & Ivoke, N. (2014). Changes in behavior and hematological parameters of freshwater African catfish *Clarias gariepinus* (Burchell 1822) following sublethal exposure to chloramphenicol. *Drug and Chemical Toxicology*, *37*(1), 107-113.

Okocha, R. C., Olatoye, I. O., & Adedeji, O. B. (2018). Food safety impacts of antimicrobial use and their residues in aquaculture. *Public Health Reviews, 39*(1), 1-22.

Plhalova, L., Zivna, D., Bartoskova, M., Blahova, J., Sevcikova, M., Skoric, M., & Svobodova, Z. (2014). The effects of subchronic exposure to ciprofloxacin on zebrafish (*Danio rerio*). *Neuroendocrinol Lett*, *35*(Suppl 2), 64-70.

Preena, P. G., Swaminathan, T. R., Kumar, V. J. R., & Singh, I. S. B. (2020). Antimicrobial resistance in aquaculture: a crisis for concern. *Biologia*, *75*, 1497-1517.

Qiu, W., Liu, X., Yang, F., Li, R., Xiong, Y., Fu, C., & Zheng, C. (2020). Single and joint toxic effects of four antibiotics on some metabolic pathways of zebrafish (*Danio rerio*) larvae. *Science of the Total Environment*, *716*, 137062.

Rahman, M. Z., Khatun, A., Kholil, M. I., & Hossain, M. M. (2017). Aqua drugs and chemicals used in fish farms of Comilla regions. *Journal of Entomology and Zoology Studies*, *5*(6), 2462-2473.

Rahman, M., Huys, G., Kühn, I., Rahman, M., & Möllby, R. (2009). Prevalence and transmission of antimicrobial resistance among *Aeromonas* populations from a duckweed aquaculture based hospital sewage water recycling system in Bangladesh. *Antonie Van Leeuwenhoek*, *96*, 313-321.

Rhee, J. S., Kim, B. M., Jeong, C. B., Park, H. G., Leung, K. M. Y., Lee, Y. M., & Lee, J. S. (2013). Effect of pharmaceuticals exposure on acetylcholinesterase (AchE) activity and on the expression of AchE gene in the monogonont rotifer, *Brachionus koreanus*. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, *158*(4), 216-224.

Rodrigues, S., Antunes, S. C., Correia, A. T., & Nunes, B. (2016). Acute and chronic effects of erythromycin exposure on oxidative stress and genotoxicity parameters of *Oncorhynchus mykiss*. *Science of the Total Environment*, *545*, 591-600.

Rodrigues, S., Antunes, S. C., Correia, A. T., & Nunes, B. (2017). Rainbow trout (*Oncorhynchus mykiss*) pro-oxidant and genotoxic responses following acute and chronic exposure to the antibiotic oxytetracycline. *Ecotoxicology*, *26*, 104-117.

Ryan, B., Medriano, C. D., Cho, Y., Kim, H., Chung, I. Y., Seok, K. S., & Kim, S. (2017). Sub-lethal pharmaceutical hazard tracking in adult zebrafish using untargeted LC–MS environmental metabolomics. *Journal of Hazardous Materials*, *339*, 63-72.

Shaaban, H., & Mostafa, A. (2023). Simultaneous determination of antibiotics residues in edible fish muscle using eco-friendly SPE-UPLC-MS/MS: Occurrence, human dietary exposure and health risk assessment for consumer safety. *Toxicology Reports*, *10*, 1-10.

Shamsuzzaman, M. M., & Biswas, T. K. (2012). Aqua chemicals in shrimp farm: A study from south-west coast of Bangladesh. *The Egyptian Journal of Aquatic Research*, *38*(4), 275-285.

Shamsuzzaman, M. M., Islam, M. M., Tania, N. J., Al-Mamun, M. A., Barman, P. P., & Xu, X. (2017). Fisheries resources of Bangladesh: Present status and future direction. *Aquaculture and Fisheries*, *2*(4), 145-156.

Shan, J., Xiaoqian, D., Xia, L., Yu, W., Zhilong, Z., Zhihui, S., & Yanjie, Q. (2022). Oxidative stress, autophagy, and apoptosis induced by doxycycline in loach fin cells in vitro. *Science of The Total Environment*, *839*, 156379.

Sosa-Hernandez, J. E., Rodas-Zuluaga, L. I., Lopez-Pacheco, I. Y., Melchor-Martinez, E. M., Aghalari, Z., Limon, D. S., & Parra-Saldivar, R. (2021). Sources of antibiotics pollutants in the aquatic environment under SARS-CoV-2 pandemic situation. *Case Studies in Chemical and Environmental Engineering*, *4*, 100127.

Vasdev, N., Deshpande, M., Katare, P., Makwana, V., Polaka, S., Tekade, M., & Tekade, R. K. (2022). New emerging technologies for genetic toxicity testing. *Pharmacokinetics and Toxicokinetic Considerations*, 175-219.

Wang, C., Lu, Y., Sun, B., Zhang, M., Wang, C., Xiu, C., & Wang, P. (2023). Ecological and human health risks of antibiotics in marine species through mass transfer from sea to land in a coastal area: A case study in Qinzhou Bay, the South China sea. *Environmental Pollution*, *316*, 120502.

Wang, H., Che, B., Duan, A., Mao, J., Dahlgren, R. A., Zhang, M., & Wang, X. (2014). Toxicity evaluation of β-diketone antibiotics on the development of embryo-larval zebrafish (*Danio rerio*). *Environmental Toxicology*, *29*(10), 1134-1146.

Yan, Z., Huang, X., Xie, Y., Song, M., Zhu, K., & Ding, S. (2019). Macrolides induce severe cardiotoxicity and developmental toxicity in zebrafish embryos. *Science of the Total Environment*, *649*, 1414-1421.

Yan, Z., Yang, Q., Jiang, W., Lu, J., Xiang, Z., Guo, R., & Chen, J. (2018). Integrated toxic evaluation of sulfamethazine on zebrafish: including two lifespan stages (embryo-larval and adult) and three exposure periods (exposure, post-exposure and re-exposure). *Chemosphere*, *195*, 784-792.

Yang, C., Song, G., & Lim, W. (2020). A review of the toxicity in fish exposed to antibiotics. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, *237*, 108840.

Yin, X., Wang, H., Zhang, Y., Dahlgren, R. A., Zhang, H., Shi, M., & Wang, X. (2014). Toxicological assessment of trace β-diketone antibiotic mixtures on zebrafish (*Danio rerio*) by proteomic analysis. *Plos One*, *9*(7), e102731.

Zargar, A., Taheri Mirghaed, A., Mirzargar, S. S., Ghelichpour, M., Yousefi, M., & Hoseini, S. M. (2020). Dietary ginger administration attenuates oxidative stress and immunosuppression caused by oxytetracycline in rainbow trout (*Oncorhynchus mykiss*). *Aquaculture Research*, *51*(10), 4215-4224.

Zhang, M., Limbu, S. M., Chen, L., & Du, Z. Y. (2021). A global analysis on the systemic effects of antibiotics in cultured fish and their potential human health risk: A review. *Reviews in Aquaculture, 13*(2): 1015–1059.

Zhang, Q., Cheng, J., & Xin, Q. (2015). Effects of tetracycline on developmental toxicity and molecular responses in zebrafish (*Danio rerio*) embryos. *Ecotoxicology*, *24*, 707-719.

Zhang, S., Ding, J., Razanajatovo, R. M., Jiang, H., Zou, H., & Zhu, W. (2019). Interactive effects of polystyrene microplastics and roxithromycin on bioaccumulation and biochemical status in the freshwater fish red tilapia (*Oreochromis niloticus*). *Science of the Total Environment*, *648*, 1431-1439.