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Pharmaceutical Drugs in Aquatic Environment and their Toxic Effect on *Pangasius* sp.: An Overview

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Abstract

One of the fastest-growing freshwater fish varieties is the *Pangasius* sp., which has a great capability for production and export growth. When *Pangasius* sp. is exposed to substances such as diclofenac, phenol, quinolones, sulfonamides, and tetracycline, even at low environmental exposure levels the fish tissue can develop chronic risk, genetic abnormalities, and histopathological changes. They come into contact with these antibiotics mostly through the discharge of pharmaceutical industry effluents, which contain antibiotic residues that are not been completely eliminated by wastewater treatment, thereby posing environmental concerns when released into aquatic ecosystems. The main objective of this review paper is to study the effect of the concentration of pharmaceutical drugs, farming techniques, and various substitutes for antibiotics that can be utilized to enhance the growth performance of *Pangasius* sp. Few studies conducted on toxicity demonstrated that increased mucus production in the gills and intestines, histological abnormalities in the liver and skin, and impaired immunoglobulin production have all been linked to sublethal phenol concentrations in fish. The consumption of these contaminated *Pangasius* sp. can raise various human health concerns. Therefore, exposure of *Pangasius* sp. to the detrimental pharmaceutical drug from industrial effluent must be held serious concern. Further research is required to conserve human health and the ecosystem.

1. Introduction

The incidence and mere presence of pharmaceuticals in the aquatic environment have become a growing concern in recent years[1]. Pharmaceutically active chemicals play a vital role in modern civilization, and they are used for a variety of objectives[2]. Many pharmaceutically active chemicals are stable and difficult to oxidize using traditional methods as they are usually lipophilic which leads to bioaccumulation[3]. Some pharmaceuticals, such as

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p-xanthine (detection frequency 0.08 %), acetaminophen (0.32%), codeine (0.16%), sulfamethoxazole (0.41%), carbamazepine (1.5%), and trimethoprim (0.08%), were detected at concentrations more than or equivalent to detection limits in a specific study[4]. Fish exposed to industrial wastewater were shown in various trials to have reproductive problems. Furthermore, fish subjected to trace doses of birth control drugs in the wide concentration range of chemical contaminants have reduced dramatically reproductive success, implying population-level effects. Unmetabolized chemicals, human medications, and their metabolites which are often released into the aquatic environment as wastewater from pharmaceutical industries, hospitals, veterinary, and sewage sludge, were observed in trace amounts. This poses a significant hazard to human and aquatic organisms' health. Pharmaceutical effluents are usually discovered to contain antibiotics, anti-inflammatory medications, antiseptics, and anti-microbials, which are toxic to aquatic life and impair water quality. At an ecotoxicological level, diclofenac has had adverse effects on numerous fish organs. Phenolic chemicals are very poisonous to people and animals, causing severe and deep damage[5]. Also, Andrieu *et al*.,[12] reported that effluents of the *Pangasius* catfish farm in Vietnam contained 0.68 µg/L of enrofloxacin and 0.25 µg/L of ciprofloxacin.

We objectively evaluate the potential repercussions of pharmaceuticals discharged into the aquatic ecosystem in this review study, focusing on the ecotoxicity that has adverse chemical and biological consequences on *Pangasius* sp., leaving human health at risk. *Pangasius hypophthalmus*, also known as *Pangasius sutchii*, is a riverine freshwater species that is one of the world's fastest-growing aquaculture and most vital inland fisheries. Striped catfish, Pla-Sawai, Patin in Malaysia, tra or basa catfish in Vietnam, iridescent shark catfish, Siamese shark, and sutchi catfish are some of the common names for the species. They are omnivorous, feeding on algae, higher plants, zooplankton, and insects[6,7]. Although *Pangasius* is frequently located in freshwater, it has an excessive tolerance for salinity, pH, dissolved oxygen, temperature, or even pollution. *Pangasius* is located to stay in salt concentrations of around 0.7% to 1% and alum water with a pH > 5 which has a tolerance for around 30°C[8].

The farming of *Pangasius hypophthalmus* has become a commercial freshwater species that is now a huge component of global whitefish supplies. On the market, the species has advanced into a completely competitive, high-cost white fish product. The aquaculture production of *Pangasius hypothalamus* is a critical enterprise in Southeast Asia and is certainly discovered in the rivers of India, Indonesia, Malaysia, Myanmar, and Thailand[7].

Pangasius sp., often known as Pangas or Basa in India, belongs to the Siluriformes order's Pangasiidae family. West Bengal and Andhra Pradesh were the primary states to execute farming in an exceedingly restricted region[6]. Later, the culturing was accomplished on an outsized scale within the states of Kerala, and Orissa. Andhra Pradesh, farms are principally located in Krishna and West Godavari districts. Small and marginal farmers were regularly growing their manufacturing of *P. hypophthalmus* throughout the country. To improve the growth performance of *Pangasius hypophthalmus*, a number of ingredients are applied throughout the culture process. Batool *et al*[44]. investigated the effects of different azomite ratios in a 38.3% crude protein diet on *Pangasius hypophthalmus* fry development, survival, and body composition.

Increased mucus production in the gills and intestines, histological abnormalities in the liver and skin, and reduced immunoglobulin synthesis have all been associated with sublethal phenol concentrations in fish, according to Vijaya *et al*[34]. Sulfonamide deposits are a major source of concern because they pose a threat to human health by establishing bacterial resistance and having negative repercussions in hypersensitive persons who are prone to allergic responses. However, sicknesses and their damaging effects on socioeconomic factors and biodiversity have expressed fears approximately long-term sustainability. The chemical residues in the *Pangasius* fish may have a long-term impact on human health.

2. Source of Pharmaceutical Drugs in the Aquatic Environment

Aquatic pollution is a major environmental problem caused by the discharge of a variety of chemical compounds that not only reduces the biological function of aquatic habitats but also cause a variety of ailments in humans[6]. The presence of detrimental substances in the aquatic environment is attributable to rising pharmaceutical consumption for human and veterinary use, as well as incorrect disposal of untreated medical waste at household locations. Wastewater treatment plants, pharmaceutical enterprises, hospital

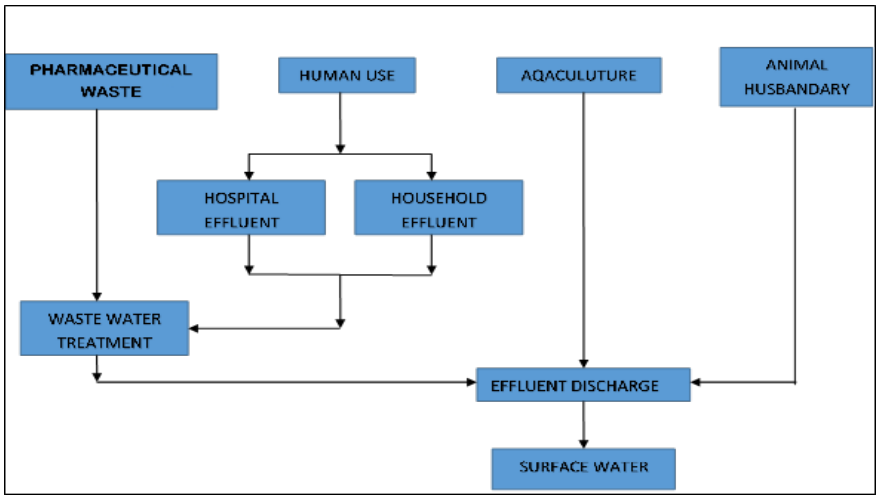


Figure 1. Entry of pharmaceutical drugs into the environment.

discharge, aquaculture facilities, veterinary usage, and other urban wastes account for the majority of these effluents[9] (Figure 1).

2.1 Veterinary Usage

Anti-infectives and hormones are active medicinal components that may be employed in both humans and animals. The factory disposal unit, as well as human APIs, are the sources of veterinary active pharmaceutical ingredients entering the aquatic environment. Veterinary APIs which are made up of urine and faeces from livestock animals are discharged directly into surface water. Tylosin is a veterinary antibiotic that has been identified in surface water. APIs are released into the environment as a result of the methods used to rear aquatic creatures, which may have adverse implications[10].

2.2 Hospital Waste

Hospital waste is thought to be the most harmful to aquatic life. Viruses and harmful bacteria, and also residual drug molecules and non-metabolized drug molecules, are among the most common pollutants found in hospital wastewater. Non-Hazardous and Hazardous wastes are two types of wastes found in hospitals. The cultivation and stock of infectious pathogens from laboratory waste, as well as surgical waste, are examples of hazardous waste. Kitchen garbage, paper plastics, blood, bodily fluids, and human parts are all examples of non-hazardous waste.

2.3 Aquaculture

The aquaculture industry is one of the largest sources of aquatic pollution due to continual effluent leaks into neighbouring water bodies. Pharmaceuticals used in aquaculture and other hazardous compounds may have negative consequences for biodiversity and other aquatic environments. Pesticides applied on agricultural grounds come to interacting directly with aquatic species, which are rinsed away by rainwater. Exposure to these pesticides has a negative impact on fish[11]. *Pangasius* mass culture is hampered by a variety of illnesses, including Aeromonas, Columnaris, Edwardsiella, and Vibrio infections[6]. In general, farmers employed antibiotics and chemical treatments to increase *Pangasius* productivity. For disinfection, these chemicals are poured straight into the water. The residual concentrations of these chemicals employed in *Pangasius* production were eventually discharged into the environment as an effluent without being properly treated, raising concerns about their possible hazardous impacts on the aquatic ecosystem around the aquaculture farms[12].

2.4 Human Medicine

Medicines are designed to cure and treat disease and to benefit human health. A few major pharmaceuticals used in human medicines are paracetamol, diclofenac, sulfamethoxazole, aminophylline, atenolol, and mebeverine[13]. Since these medications are not destroyed

after treatment, a part of them may be released into the aquatic environment, potentially harming the ecology. Pharmaceuticals are typically assumed to have no detrimental environmental impact, yet drinking water as well as eating fish that have accumulated pharmaceutical residues could be harmful to humans[14].

2.5 Urban Waste

In recent decades, the world has seen massive urbanization and industrialization. Although a large number of sewage treatment plants and wastewater treatment facilities comply with regulatory requirements, unmetabolized medicines are nonetheless released into the environment, particularly in rivers, causing water contamination[15]. Wastewater from urban runoff and industrial discharge degrades water sources and harms public health[16]. According to the United Nations, 80 per cent of untreated wastewater is currently released into aquatic bodies. More than 50 pharmaceutically active compounds have been detected inside the aquatic environment[17]. Drugs including diclofenac, fluoroquinolones, oxytetracycline, and sulfamethoxazole have been found in water bodies. The therapeutic chemical diclofenac was identified in rivers and streams in Switzerland[18]. Antibiotics like ciprofloxacin and fluconazole have been found in exceptionally high amounts in India. To protect freshwater resources, extensive river water monitoring is becoming increasingly important[15].

3. Chemical Compounds Exposed to *Pangasius* sp.

Antibiotics, parasiticides, heavy metals, disinfectants, probiotics, and other food supplements have become essential components in the prevention and treatment of bacterial and parasitic diseases. These substances have aided the growth and productivity of the Asian aquaculture sector, but they have also drawn criticism. Chemical compounds in cultured organisms pose a risk to human consumers and highlight the limitations of food safety controls. They also cause an ecological imbalance by modulating a few processes like denitrification, nitrogen fixation, etc., which affect the fish's physiological functions. Chronic toxicity and histological changes in fish tissue have been reported as a result of these antibiotics' negative impacts[21].

3.1 Quinolones

The extensively observed antibiotics in the aquatic environment are said to be quinolones in conjunction with sulfonamides. Quinoline incorporates the fluoroquinolone family as they own fluorine atoms of its chemical structure. Fluoroquinolones in human medicine consist of enoxacin, ofloxacin, sparfloxacin, temafloxacin, and tosufloxacin. They consist of antimicrobial properties which can be probably beneficial in veterinary medication. Few fluoroquinolones collect resistance at some point of scientific use in humans.

Enrofloxacin (ENR) belongs to the fluoroquinolone family which has been established to be extremely beneficial against bacterial infection in fish and is widely adopted in culturing of Tra catfish (*Pangasianodon hypophthalmus*) in Vietnam[22]. In *Pangasius* farms, the vastly used antibiotic was found to be enrofloxacin, which has a high drug concentration in effluents[23]. In *Pangasius* catfish farms within the Mekong Delta, Vietnam, the effluent exhibited the most attention of 0.68 µg/L concentration for enrofloxacin. About 18% of the enrofloxacin mass is discharged into the neighbouring aquatic environment[12]. Enrofloxacin and its metabolite ciprofloxacin may also rapidly cause oxidative strain that may be dangerous to the organism[22]. Enrofloxacin has been utilized in catfish farming to treat Bacillary Necrosis of *Pangasius* [BNP]. ENR and its metabolite ciprofloxacin in striped catfish are removed in medicated feed under tank situations 98-188 µg ENR/kg and 13-17 µg CIP/kg in muscle tissue (inclusive of skin) seven days after the remedy[24].

3.2 Sulfonamides

Vietnam is the third-largest *Pangasius* production country in the world. Several bacterial and parasitic infections have accompanied *Pangasius* production. Antibiotics are commonly applied to promote the growth of fish or prevent infectious diseases[25]. The function of sulfonamide is to inhibit bacterial synthesis of dihydrofolic acid and protein. Sulfonamides are used within the post-larvae to the adult stage[26]. Liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS) is an extensively used approach for the one-time analysis of the presence of sulfonamide[25]. Analyzes of four Sulfonamide Antibiotics (SAs) *viz*. Sulfamethoxazole (SMX), Sulfamethoxydiazine (SMD), Sulfamethazine (SMT), and Sulfadiazine (SDZ), may be executed with the aid of using the LC-MS/MS

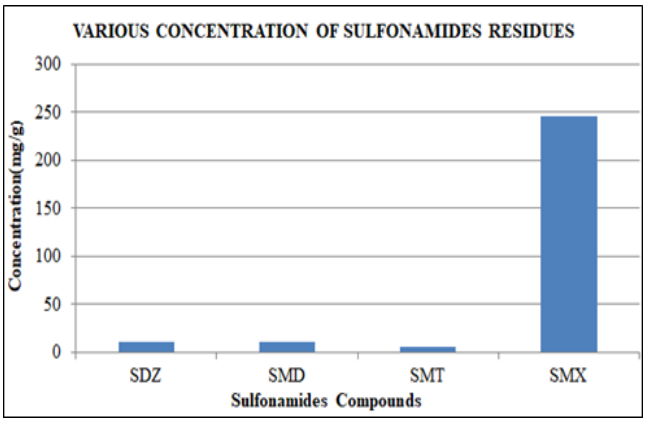


Figure 2. Highest concentration of sulfonamide residues attained in *Pangasius*.

technique. Due to its low biodegradability, SMX can remain in the environment for long periods. In seawater, SMD and SMT were present at low concentrations[27]. The maximum degree of SMX attention in *Pangasius* fish is 245. 91 µg/kg. The highest concentration of sulfonamide residues in *Pangasius*[28] is shown in Figure 2.

3.3 Phenol

Phenol is a colourless-to-white solid with a characteristic odour. Phenol has been named on the top 20 list of chemicals likely to pose the highest risk. Phenol is extensively used in different chemical industries[29]. The main toxic components of industrial wastewater were phenolic compounds. Consumption of water containing a higher concentration of phenol is known to cause various detrimental effects on humans[29]. Phenolic compounds are classified into four main groups: Flavonoids, phenolic acids, lignans, and tannin polymers. Phenol enters into the blood circulation of fish through gills and mucous epithelium of the mouth and accumulates into different organs[30]. The sub-lethal concentration of phenol at 2.85 and 4.11 mg/L is more injurious to the aquatic ecosystem. The presence of phenol in the water bodies has toxic effects on the *Pangasius* sp. and disturbs their oxidative stress and antioxidant balance[31].

3.4 Tetracycline

Antibiotics are used to protect fish from bacterial disease and as well as growth promotion. Tetracycline antibiotics are released into the environment through urine and faeces from humans and animals. Predominantly a group of these antibiotics such as oxytetracycline is used as an antibacterial against *Clostridium difficile* in aquaculture production. Oxytetracycline (OTC) is from tetracyclines class, used to treat contaminations of the respiratory and urinary lots, skin, ear, eye, and gonorrhea[32]. It has a high absorption capability in terrestrial and aquatic environments[3]. The concentration of tetracycline was found to be below 1 mg/L in the aquatic environment[33].

3.5 Diclofenac

Diclofenac is a significantly active drug compound in surface water samples. It is a non-steroidal, anti-inflammatory drug that by default prevails in the aquatic environment. Diclofenac is utilized in the form of capsules, ointments, gels, and tablets for skin application. It fundamentally diminishes aggravation and firmness because of rheumatoid joint pain, osteoarthritis, and stomach cramps identified with mensuration and spondylitis. It causes ringing in the ears, migraine, obstruction, and gas or bulging in people because of the utilization of diclofenac. Diclofenac drug causes expanded danger of liver harm. In the wake of devouring the diclofenac drug for a half year, it caused liver sickness in 85% of individuals[34].

3.6 Metals

Heavy metals such as Cadmium (Cd), Nickel (Ni), Chromium (Cr), and Lead (Pb) were tested in various organs such as gills, livers, kidneys, and flesh tissues of control fish living in natural water systems. The majority of the metals are found in edible portions of fish. Eating fish has an impact on humans as well, causing a number of health issues. Lead toxicity has become a major concern due to the dangers it poses to human health. Lead enters water systems *via* runoff, industrial waste, and sewage waste. Pb exposure can also cause reproductive problems, osteoporosis (brittle bone disease), and high blood pressure[19].

4. Farming Techniques

Extensive pangas farming generally uses an improved version of conventional methods, along with low stocking density and low inputs, such as feed, fertilizer, labour, and management. Stocking and also other inputs are used at intermediate levels in semi-intensive operations. The intensive production process is characterized by a high stocking level and a high input level. Panga farming

Table 1. Different research on the effects of various chemicals on the *Pangasius* sp

Organisms	Compounds and Method Used	Study Conducted	Reference
Pangasius catfish	Enrofloxacin, Sulfamethoxazole; LC/MS-MS	About 90 samples of imported *Pangasius* catfish were purchased from Thailand. Antibiotics were extracted from the catfish samples using a method developed by Dasenaki and Thomaidis for extracting antibiotics from tissue. Enrofloxacin residue was discovered in 9 non-skin and 5 skinned *Pangasius* catfish samples. Researchers discovered veterinary antibiotic residues in *Pangasius* that pose a health risk using Hazard Quotient (HQ). The hazard quotient represents the health risk posed by eating imported Thailand *Pangasius* catfish. When the HQ value is equal to or even less than one, no major risk to human health exists. Skin samples of *Pangasius* catfish had higher enrofloxacin concentrations than non-skin samples, and they were also higher than the average level of enrofloxacin contamination in this study (7.5 g/kg).	23
Pangasius species	Diclofenac; HPLC	*Pangasius* Sp. were purchased from Andhra Pradesh's coastal districts. Sharks (*Pangasius*) were exposed to varying doses of diclofenac after acclimating to the surrounding environment. The LC50 was measured at 264 mg in 11 litres of diclofenac-exposed fish. The liver tissue samples of the control group fish and the dead fish after exposure to diclofenac were taken for histopathological analysis. Histological changes in liver tissues were examined The HPLC was done using a modified Dubois method. HPLC analysis, the chromatogram of control fish liver tissue showed no significant changes. Diclofenac-exposed tissue had a peak value of 0.24 AU. These findings matched those of Vijaya's previous study on the negative effects of diclofenac in *Pangasius* Sp.	34
Pangasianodon hypophthalmus	Enrofloxacin, Ciprofloxacin; LC/MS-MS	The focus of this research, according to Wang *et al.*,[22] research, is to see how enrofloxacin (EF) affected Tra catfish, *Pangasianodon hypophthalmus*, in terms of density stress. *Pangasianodon hypophthalmus* was available from commercial fish farming in the Mekong river. Also, the research described a method for determining Liquid Peroxidase (LPO) concentrations in the gills, brain, and liver (2000). With various modifications of the previous methods practised, Catalyse (CAT) activity in the gills, brain, and liver was measured. The Habig technique was used to measure Gluthatione S-Transferase (GST) activity in the gills, brain, and liver. GSH levels in the gills and liver were determined. A calibration curve was created using pure Gluthatione levels (GSH) serially diluted in SSA (1 per cent) solution to determine GSH concentration. Activity tests (AChE) were measured in the gills, brain, liver, and muscles. CAT activity was significantly higher in EF fish raised at a medium density.	22

Pangasius species	Phenol, clofibrate; RP-HPLC	Shark fish (*Pangasius*) were exposed to various concentrations of phenol and colfibrate in a previous study. *Pangasius* spp. was raised in Andhra Pradesh's coastal regions. A modified Dubois *et al.*, procedure was used for the RP-HPLC. To determine lipid peroxidation activity, the TBARS test was used. The catalytic activity of several oxidative stress indicators was assessed using a Spectrophotometric approach. The Total Antioxidant Potential was calculated using the phosphomolybdenum technique. In response to exposure to both chemicals, the total antioxidant capacity of fish hepatocytes revealed that phenol was more harmful than clofibrate.	41
Pangasianodon hypophthalmus	Quinolones, Sulfonamides, Tetracycline; UHPLC-MS/MS	40 blank *Pangasius* fillet samples were purchased from Colpani Pisciculture in Mococa, Brazil, as a key component of the Bortolotte *et al.*,[27] studies, with the assurance that the fish had not been exposed to the veterinary medications that were the subject of this study's analysis. Only the muscle was utilised as a blank sample during the verification and application of the analytical procedure. On an Agilent Zorbax SB C18 (3.5 m, 4.6 mm, 150 mm) column in a UHPLC 1290 system, chromatographic separation was performed. Validation was done in accordance with the European Community's and the Brazilian Ministry of Agriculture, Livestock, and Supply's guidelines. This report looks at precision and accuracy, Limit of Detection (LOD), Limit of Quantitation (LOQ), and resilience as validation characteristics. Anastassiades' QuEChERS method was used for extractions during sample preparation.	27

takes place from March to December. Most farmers stock their ponds from March to May and harvest the fish after three months[35]. Ninety-two per cent of farmers in India cultured *Pangasius* with other fish (polyculture), whereas only eight per cent did so with carp. Carps were used by the majority of polyculture farmers, including 10% rui, 6% silver, and 5% mrigal[36]. Catfish larval rearing at the nursery level is hindered by high mortality, resulting in inefficient production. Probiotics based on mixed Bacillus minimize *Pangasius* mortality and can be used as a natural antibiotic during the first phase of rearing by hatcheries and nurseries[37]. *P. hypothalamus* is a riverine fish that matures after three years. Males and females are easily distinguished; females are distinguished by their large, swollen, soft, and reddish-pink-bloated bellies, while males are differentiated by their reddish genital openings and milt oozing[38]. In Vietnam, two types of feed are used in catfish farming: Manufactured Pelleted Feeds (MPF) and Farm-Made Feeds (FMF). The MPF is floating feeds that are prepared on-site, whereas the FMF are sinking feeds that are prepared on-site[39]. *Pangasius* sp. is highly resistant to pH, salinity, dissolved oxygen, temperature, and even pollution, making it a good source of calories and protein for poor, middle-class, and upper-class people in both urban and rural areas. *Pangasius hypophthalmus* is well-liked by a wide range of people, making it a valuable food source for many communities all over the world[36]. Pangus farming has begun to decline, and farmers have identified a number of issues, including high input costs and a low market price. It is necessary to upgrade pangas farmers' management practices in order to improve their socioeconomic conditions[40].

5. Methodology

Few studies on *Pangasius* sp. have been performed to estimate the impact of antimicrobials and the biological changes that occur as a consequence of the prevalence of pharmaceutical drugs in the aquatic environment. Various methodologies were used to investigate the

physicochemical and biological effects of toxins on *Pangasius* sp. (Table 1). When *Pangasius* sp. is exposed to substances like diclofenac, phenol, quinolones, sulfonamides, and tetracycline, even at low environmental exposure levels the fish tissue can develop chronic risk, genetic abnormalities, and histopathological changes.

6. Growth Performance and Bioremediation

Due to their rapid development, ease of culture, excellent disease resistance, and wide range of environmental conditions, *Pangasius hypophthalmus* and *Pangasius* are attractive fish species for prospective and progressive fish aqua culturists[43]. A variety of components are used during the culture of *Pangasius hypophthalmus* to ameliorate the growth performance. The recent research conducted by Batool *et al*.,[44] looked at the effects of varied azomite ratios in a 38.3% crude protein diet on *Pangasius hypophthalmus* fry growth, survival, and body composition. The trial fish were separated into three treated groups (levels 0 % control, 0.5 % T1, and 1.0 % T2), after four weeks of colonization and isolation treatments. T2 fish had substantially greater growth indices (P=0.05) than T1 or control fish, and their length increment was also significantly varied. The relevance of trace elements as vital constituents in diets is also obvious in fish, albeit in modest amounts. They demonstrated that a 1% azomite addition to the fish protein can substantially improve development without influencing the biochemical nutritional profile of the fish[44]. Following this research, Haque *et al*.,[37] conducted a study adopting mixed *Bacillus* derived probiotics to mitigate *Pangasius* fry mortality and use it as a sustainable substitute to antibiotics during the first phase of fish growth and production. They examined how probiotics affected *Pangasius* fish growth, hematological markers, morphological changes and gut microbial content. The trials were divided into three phases: larvae to fry (20 days), fry to fingerlings (45 days), and fingerlings to marketing (90 days). Treatment groups had considerably (p 0.05) greater survival percentages in phases 1 and 2. As a result, the addition of probiotics resulted in a positive modification in the morphological characteristics of the intestine.

Furthermore, when the larvae were fed six times a day, Vu N *et al*.,[45] found that they had a high survival rate of 66%. In the preliminary extrinsic diet, the *P. hypophthalmus* larvae were fed with *Brachionus angularis* rotifers for the first three days and afterwards, *Moina macrocopa* water fleas for the next seven days. Earlier feeding of live *B. angularis* rotifers to *P. hypophthalms* larvae mitigated starvation and cannibalism. The growth performance of silver catfish larvae was hampered by a delay in their initial feeding time.

Another research indicated that lipids had a protein-sparing impact, as fish fed a 30 per cent protein/12 per cent lipid diet grew and used feed similarly to those fed a 40 per cent protein/12 per cent lipid diet. Variations in lipid and protein feeding had a significant impact on specific growth rate (SGR) and Feed Conversion Ratio (FCR) diet, a 40% protein/12% lipid diet, had the highest SGR. When dietary protein was elevated from 25% to 30%, the HSI dropped, but then increased modestly. The 40/12 diet had the least FCR, and only the 25/6, 25/12, and 30/6 treatments had higher FCRs[46]. When fish were fed three different levels of crude protein, they showed superior development in t3 (40 per cent CP) with declining enzyme production, according to Khan *et al*[43]. Overall, fish fed 40% CP feed performed much better, with no negative impacts on the nutritional content of the fish. RBCs and WBCs were profoundly influenced by CP levels up to 40%, but when protein source levels increased; their numbers began to fall to 44%. The amount of dietary protein had a favourable effect on fish Hb levels, with no significant differences (P 0.05) between the 44 per cent and 40 per cent CP diets. Thi Da C *et al*.,[47] conducted a similar experiment to assess the growth performance of striped catfish. Fish meal is the principal source of Crude Sietary Protein (CP). CP made from local resources has been used to replace 20 to 100 per cent of the fish feed CP. The findings imply that dietary protein can be substituted with protein derived from locally accessible plant and animal sources without affecting growth or carcass characteristics. The golden apple snail meal regimen had the greatest intra-peritoneal fat index (IPF per cent), followed by the shrimp head meal and soybean meal diets. The kidney index ranged from 0.4 to 0.7 and was unaffected by therapy.

Artemisia pallens is a medicinal plant with an aroma. A study investigated by Shreenidhi *et al.,* 2021 has identified it as a protective substance against oxidative damage. The presence of phytocompounds such as flavonoids, phenols, terpinols, and others in *Artemisia pallens* has been linked to its hepatoprotective activity. *Artemisia pallens* reduces hepatic cell inflammation and improves liver function in fish[48].

Spirulina had long been suspected to be an immunostimulant. It activates non-specific immune systems to supplement the elements of the mucosal and systemic immune systems. Other medications such as phenol and clofibrate, and also diclofenac, have a misconstruing impact in conjunction with the histopathological cell rupture. The therapeutic effects were observed once the algae spirulina was introduced within the cells, inside the instance of liver cells, as the algae began to justify constrained vacuole spaces in comparison to the chemicals exposition. The reduction in vacuole formation was also configured and had a better effect in the intestinal tissues by the inclusion of Spirulina[49].

7. Toxicity of Individual Compounds on *Pangasius* sp.

The toxicity study conducted by Vijaya *et al.*, (2018) demonstrated increased mucus production in the gills and intestines, histological abnormalities in the liver and skin, and impaired immunoglobulin production linked to sublethal phenol concentrations in fish. The diclofenac-exposed fish's hepatocytes were slightly larger, with significant lesions. Changes in blood pressure (one of the diclofenac's side effects) caused abnormalities in the liver's red blood cells[34].

Sulfonamide deposits are a serious supply of concern as a result they create a risk to human health through the event of bacterial resistance and harmful consequences in super sensitive people, who adore allergic reactions[50]. According to Wang *et al.*,[22] when the *Pangasianodon hypophthalmus* was exposed to Enrofloxacin (ENR), the digestive tract, which is the primary route of absorption, and the gills were contaminated as a result of ENR blood transport. The detoxification of Enrofloxacin (ENR) in *Pangasiandon* appears to lead to an excess of Reactive Oxygenated Species (ROS), which fails to balance the cycle in the gill but does not cause harm to the brain and liver. Environmental stress, such as rearing density, can be used to determine physiological responses to ENR. When the fish was raised at medium density (80 fish/ m^3) appeared to bear easily with the ENR treatment, whereas fish rose at low (40 fish/ m^3) and high density (120 fish/ m^3) and did not (120 fish/ m^3). As a result, these findings suggest that various density levels may potentially function as a stressor in *Pangasianodon hypophthalmus* .

Bacillus sp. might play a beneficial function in eliminating waste materials from aquaculture habitats, maintaining optimum quality of the water, and lowering the stress of susceptible aquatic species, all of which can contribute to improved immuno-physiological balance, increased survival and better growth. A lower dosage (105 CFU per mL) containing *B. licheniformis* Dahb1 was more successful at improving the production of catfish (*Pangasius hypophthalmus*) than a larger dosage (107 CFU per mL)[51].

Paracetamol (APAP) causes severe liver harm in fish, mice, and humans, according to a study. The exact mechanism of APAP-induced liver toxicity is unknown; however, the reacting metabolite NAPQI is known to be involved. It also implies that APAP and its other volatile metabolites, along with oxidative stress metabolites, may interact with APAP drug targets such as Gpx and GST. Elevated levels of protein carbonyl TBARS and LHP in freshwater fish liver tissue indicate APAP-induced protein and lipid degradation[52].

8. Effect of Pharmaceutical Drugs in Human Health

Antibiotic usage in *Pangasius* catfish production has the potential to contaminate the fish through the

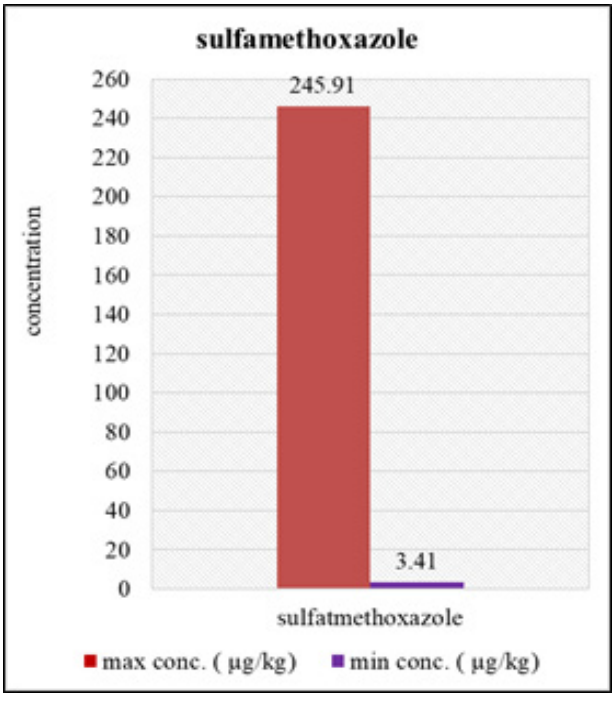


Figure 3. Sulfamethoxazole concentration in *Pangasius*.

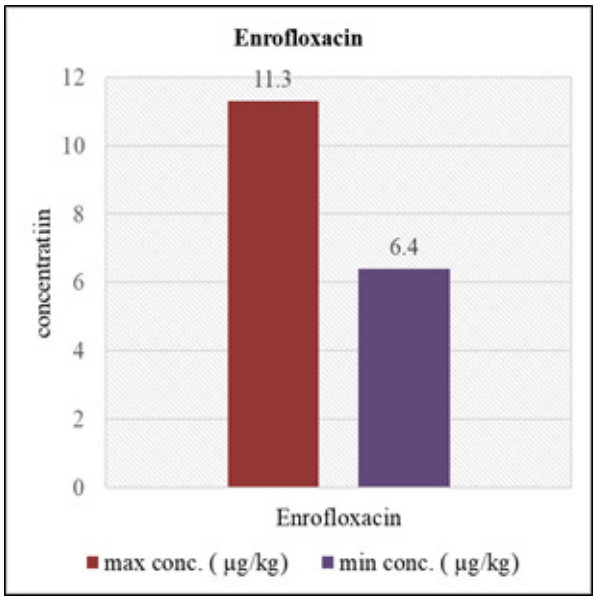


Figure 4. Enrofloxacin concentration in *Pangasius*.

bioaccumulation of antibiotic residues of concern in fish tissues. Antibiotics are inadvertently ingested by people who consume these goods. The results of the Jansomboom *et al.*,[23] reports on Sulfamethoxazole (Figure 3) and enrofloxacin (Figure 4) which were found in one of the samples, show that the health risk from *Pangasius* catfish consumption associated with those two antibiotics was insignificant if per capita intake rate of *Pangasius* catfish

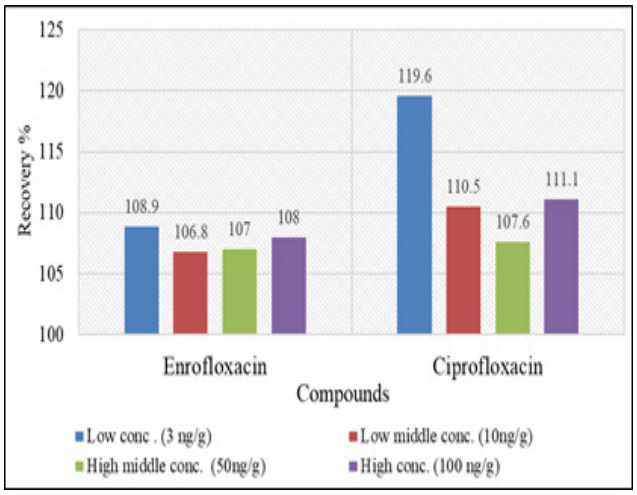


Figure 5. Accuracy of the method for the quantitation of quinolone residues in *Pangasius* fillet by HPLC-MS/MS.

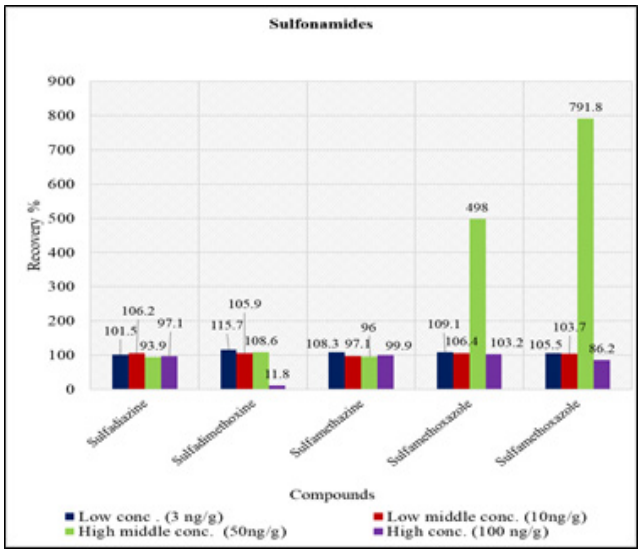


Figure 6. Accuracy of the method for the quantitation of sulfonamides residues in *Pangasius* fillet by HPLC-MS/MS.

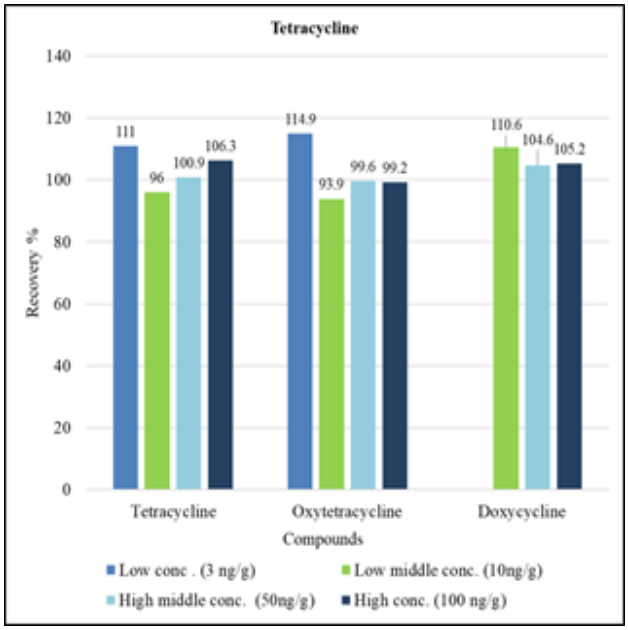


Figure 7. Accuracy of the method for the quantitation of Tetracyclines residues in *Pangasius* fillet by HPLC-MS/MS.

products is 62.40 g fish per person per day (the rate of catfish consumption in Thailand).

The accuracy of each compound was assessed by the recovery test in Bortolotte *et al.*,[27] reports by HPLC-MS/MS. The graphs above depict the quantity of antibiotic residue recovered from *Pangasius* fillets at various concentration levels[27]. We can deduce from this research that a particular percentage of antimicrobial residues (enrofloxcin and ciprofloxacin) are recovered from *Pangasius* fillets at four levels of concentration. In the low concentration, enrofloxacin and ciprofloxacin had the highest percentage of recovery (Figure 5). The sulfonamide compounds sulphamethoxazole and sulfathiazole exhibited the highest recovery percentages at high-medium concentrations, while the recovery percentages of the other compounds were nearly identical at all concentration levels (Figure 6). However, doxycycline had no recovery percentage at low concentrations in tetracycline, whereas oxytetracycline had the highest recovery % (Figure 7). The existence of antimicrobial residues might pose a danger to human health[27].

9. Conclusion

Pangasius, a widely consumed fish, is becoming endangered due to human pollution. This could have a significant economic impact as India is a major aquaculture producer around the globe. More than 10% of the world's fish diversity is in India. In fish, enrofloxacin detoxification appears to cause an increase in Reactive Oxygen Species (ROS). *Pangasius* appears to struggle to balance this detoxification in the gills, but it appears to have far less of an impact on the liver and brain. Muscle difficulties can also be caused by synapse disorders. Environmental stressors such as rearing density, on the one hand, and the physiological reaction of fish to enrofloxacin, on the other hand, may have an impact on the physiological response of fish to enrofloxacin. Periodic checks in water bodies can help to reduce the addition of contaminants or chemicals. Even if the concentrations of chemicals/pollutants are very modest in some circumstances, there is a considerable risk of organism death when these molecules aggregate. Bioremediation like the addition of Bacillus derived probiotics and Spirulina had a positive effect on the morphological characteristics of the intestine and enhanced the growth performance. Enrofloxacin residues in *Pangasius* should be avoided by competent authorities. As a result, *Pangasius* exposure to antibiotics and other chemical compounds must be prioritized, and pollutants or chemical addition can be reduced by repeating water quality assessments.

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