10. Study on Impact of Aquaculture Drugs and Chemicals on Fish Health, Productivity and Biodiversity

10.1 Executive Summary

Ten different categories of chemicals were found to use in aquaculture activities. They are antibiotics, disinfectants, gas removal, oxygen supplier, vitamins and minerals, growth promoter, insect killer, algae killer, predator killer and pH balance. In pharmacy of investigated areas, 50 different types of aquaculture drugs and chemicals were recorded. Among those, 15 types are widely used by the farmers such as Renamycin, Amoxifish, Ossi-C, Timsen, Aquamysine, Aquamycine, Virex, Aquakleen, Geolite gold, Oxy Dox F, Polgard plus, Charger gel, Seaweed, Bactisal and Deletix. Productions of Pangas and Koi in Gouripur and Muktagacha Upazillas were almost double in the chemical treated ponds compared with non treated ponds. However, Pangas production in Muktagacha was almost double than Gouripur, where stocking density was also higher. Whereas, in BAU experimental ponds, production of Pangas was significantly higher in the non-treated ponds compared with chemical treated ponds. On the other hand production of Koi was almost similar in the treated and control ponds.

In Upazillas like Fulpur, Muktahacha and Fulbaria EUS, dropsy, Edwardsiellosis diseases were recorded with Pangas and Tilapia. Shing had 90-100% mortalities within very short period from unknown diseases with no obvious symptoms (except swollen abdomen and spots) provided 30-100% recovery after application of drugs. In EUS affected Tilapia in Fulpur (20%) and Muktagacha (30%) and farmers used Renamycin, Polgard plus and Ossi-C with a result of 80-95% recovery. In Edwardsiellosis affected Pangas in Fulpur (80%) and Fulbariaa (50%), farmers used Renamycin, Timsen, Polgard plus and Ossi-C in Fulpur and Geolite and Timsen in Fulbaria having 80% recovery in both Upazillas. Farmers used Aquamycin and Ossi-C against dropsy in Fulpur with a result of 95% recovery. Zoothamnium and various spots on skin, scale loss in Koi, where farmers used Renamycin, Aquamycine, Ossi-C and Polgard plus drugs as treatments and achieved 70-80% recovery. It was thus observed that aqua drugs played excellent role in recovery of fish diseases and maintenance of health. In a drug treated pond of BAU, plankton bloom was recovered within one week by seaweed (Algicide) application. In laboratory study, clinically all the fishes of various regions did not show any remarkable changes in

chemical treated and non-treated ponds. Histopathology in the control ponds, skin-muscle, liver, kidney and gill of fish had almost normal structure. However, in the chemical treated ponds, the above mentioned investigated organs of fishes had remarkable pathological changes like necrosis, hemorrhage, vacuum, melanocytes and partial loss of organs. Thus fishes of chemical treated ponds although clinically looked normal, but histopathologically they were severely affected. It could be mentioned that, drugs and chemicals had some adverse effect on the health of culture fishes.

10.2 Background and Justification

Aquaculture is the fastest growing food-producing sector in the world. It is growing more rapidly than all other animal food producing sectors. Aquaculture in Bangladesh is also under heavy expansion. Over the last decade it has expanded, diversified, intensified and technologically advanced. In aquaculture, the external inputs required for successful fish production is chemical, which has been used in various forms for centuries (Subasinghe *et al.* 1996). They are essential components in pond construction, health management, soil and water management, enhancement of natural aquatic productivity, transportation of live organisms, feed formulation, manipulation and enhancement of reproduction, growth promotion and processing and value enhancement of the final product (GESAMP 1997, Subasinghe *et al.* 1996). Intensification of aquaculture brings about the use of more chemicals and antibiotics in this sector.

Antibiotics have been applied in aquaculture for over 50 years for treating bacterial infection in fish. Since then, their use has grown both in numbers and quantity, as the problem of bacterial disease has increased (Inglis 1996). Bacterial disease occurs most frequently and severely in intensive culture systems and it is there that most antibacterials are used. Use of antibiotics in aquaculture may contribute to increased resistance and cause detrimental effects in medicine more generally. A variety of other chemicals are also used in aquaculture for health management of fish apart from antibiotics. Some common chemicals include sodium chloride, formalin, malachite green, methylene blue, potassium permanganate, hydrogen per oxide, copper compounds, glutaraldehyde and trifluralin (Plumb 1992).

A range aquatic disease could be found in farmed aquatic animals of Bangladesh (BFRI 1999, Faruk *et al.* 2004). Epizootic ulcerative syndrome (EUS) in fish and white spot syndrome in cultured shrimp are the two most devastating diseases have huge impact in aquaculture of Bangladesh. Farmers are using a range of chemicals and antibiotic for the treatment of diseased animal. Also, pharmaceutical companies and chemical sellers are influencing fish and shrimp farmers to buy their products. It has been realized that farmers have been using these chemicals without knowing their effectiveness.

There are problems associated with the use of chemicals. Excessive use of antibiotics for disease treatment can lead to development of resistant strains of bacteria as well as consumer boycotts in importing countries due to residues of antibiotics. EU countries are concerned about the use of hazardous chemicals in fishery products especially in shrimp and prawn products. Rejection of consignment by European Commission's is quite common due detection of nitrofuran in prawn bodies. Nitrofuran antibiotics are veterinary drugs whose use in food-producing animals and fish is banned in the EU because of health concerns, including a possible increase in cancer risk in humans.

However, there is lack of information on the use of chemicals and antibiotics in aquaculture industry of the country and the impact that they have on fish productivity and biodiversity and needs examination. This project would address some of these issues.

10.3 Literature Review

In a recent study Faruk *et al*. (2008) found a range of chemicals including antibiotics used in aquaculture for fish health management and disease treatment. Along with commonly used traditional chemicals, they found a number of new products with various trade names like JVzeolite, Geotox, Green zeolite, Orgavit aqua, Fish vitaplus, AQ grow-G, Oxy flow, Oxy max and O_2-marine were the most widely used compounds. Fourteen branded antibiotics were found with different trade names for disease treatments of aquatic animal. Major active ingredients of these antibiotics were Oxytetracycline, Chlorotetracycline, Amoxicillin, Co-trimoxazole, Sulphadiazine and Sulphamethoxazole. They reported thirty three pharmaceutical companies were seen active for producing and marketing of these products. Some of these products have been marketed by different companies from the countries like India, Thailand, Taiwan, Indonesia, Malaysia, Spain and USA.

Faruk *et al.* (2005) observed that commonly used chemicals in aquaculture are lime, salt, urea, triple super phosphate (TSP) potassium permanganate, vitamins, antibiotics (mainly oxytetracycline and chlorotetracycline), rotenone, phostoxin, sumithion, melathion and some hormones. They also stated that most of the farmers used chemicals and antibiotics indiscriminately without knowing their mode of action, doses and appropriate procedures of application. Sarker (2000) conducted an experiment to test drug sensitivity of five isolates of *Aeromonas sorbia* and found that most of the isolates were sensitive to Oxytetracycline (OT), Oxolinic acid (OA) and Chloramphenicol (C) but resistance to Erythromycin and Sulphamethoxazole (SXT).

A variety of chemicals are used in aquaculture for health management of fish apart from antibiotics. Some common chemicals include sodium chloride, formalin, malachite green, methylene blue, potassium permanganate, and hydrogen per oxide, copper compounds, glutaraldehyde and trifluralin (Plumb 1992). Sodium chloride is an old treatment used for a variety of diseases of fish. It is especially effective chemical when treating some fungal and parasitic diseases in fish. Formalin is versatile compound used in a variety of ways in treating fish. Formalin is used primarily as an external parasiticide on fish and fish eggs as either flush, prolonged, or indefinite treatment for fungus control. Potassium permanganate ($KMnO_4$) is one of the widely used chemical in fish health management. It is a strong oxidizing agent approved for the purpose to treat ponds. Potassium permanganate is good for treating external protozoa and external bacterial infections (Plumb 1992). Malachite green is an organic dye that has been popular as a parasiticide and fungicide on fish. It is principally used in hatcheries rather than grow-out systems. Lengthy withdrawal period is essential following application because of persistent residues (Alderman 1992).

Concern is growing over the use and potential misuse of some of aquaculture chemicals. The amount of information on chemical use in aquaculture and its significance for human health assurance, environmental protection and sustainable development of the sector, has been increasing throughout the last two decade (FAO/NACA 1995, Plumb 1995). Pesticides are also used in aquaculture of disease treatment, such as organophosphates, organotin compounds, rotenone and saponin, dichlovos, trichlorfon, diptarex, melathion, dursban are the widely used organophosphate applied to control ectoparasitic crustacean infections in finfish culture. For all the organophosphates, effects on non-target organisms, particularly crustaceans are major concerned. Due to high neurotoxicity of organophosphates, potential effects on health of fish farm workers are also health hazardous chemical (Alderman *et al.* 1994).

When chemicals are employed by aqua culturists, either for preventative or treatment purposes, a certain portion of the applied substance leaves the farm via the effluent or, in the case of pond culture system or net-cage culture, is released directly to the environment. Cravedi *et al.* (1987) reported that the vast majority of Oxytetracycline and oxolinic acid provided is likely to leave the farm as particulate wastes because of feed wastage and poor digestive absorption of these drugs. Discharge of these contaminated feeds and faeces is likely to occur continuously at low concentrations but may be greater at certain periods of the production cycle such as during tank or pond cleaning. Accumulation of solid wastes and associated chemical residues near the point of discharge is likely.

With the expansion of aquaculture in Bangladesh, there has been increasing trend in using more chemicals in aquatic animal health management. Commonly used chemicals in Bangladesh aquaculture are lime, rotenone, various forms of inorganic and organic fertilizers, phostoxin, salt, dipterex, antimicrobials, potassium permanganate, copper sulphate, formalin, sumithion and melathion (DoF 2002, Faruk *et al.* 2005).

10.4 Objectives

The objectives of the present study are:

i. To compare fish production between culture systems using chemicals and without chemicals

ii. To study of impact of use of chemicals on fish health and productivity

iii. To identify problems associated with their use

10.5 Experimental design and Methodology

Study area: Muktagacha, Fulbaria, Fulpur and Gouripur Upazillas of Mymensingh district were considered as study area for collecting data on use of aqua-drugs and chemicals used in freshwater aquaculture.

Data collection: Secondary data on the use of chemicals and antibiotics were collected with GO and NGO offices. The primary data were collected through the combination of the following survey techniques**:**

Questionnaire survey: Fish farmers, hatchery and nursery owner, farm workers, chemical retailers, and other stakeholders were questioned individually. A set of preliminary questionnaire based on the objectives of the study was prepared. Major topic of questionnaire were the name of chemical, active ingredients, purpose of use, method of application and dose, duration, source, effects on environment, price, impact on health and productivity. In addition data on farming practices, general farm management, health and disease problems, seasonality, mortality etc were also gathered. For the interview, simple random sampling method will be followed during interview.

Laboratory analysis: Laboratory study was focused on the verification of fish health and diseases of aquatic animal reported by farmers and the assessment of the efficacy of potential selected chemicals and drugs in curing disease in laboratory and field condition.

Health and disease issues: In addition with the field observation, diseases of fish were verified through laboratory analyses. Fish were examined clinically through observing gross signs, abnormality, lesions and erosions and external parasites. Samples for histopathological examination will be taken from skin, muscle, gill, liver and kidney, and were fixed in 10% neutral buffer formalin. The samples were processed in an automatic tissue processor, stained with haematoxylin and eosin, mounted with Canada balsam and will be examined under compound microscope.

Impact assessment: Fish production between culture systems using chemicals and without chemicals were compared. For this purpose, four possible culture systems like Thai Pangas, Thai Koi, Tilapia and Shing were chosen and total fish production and water quality were determined from ponds using chemical and without chemicals. This experiment was conducted in experimental ponds at BAU. The health status of fishes and production were also compared with Thai koi and Thai pangas.

10.6 Experimental design in BAU ponds

Eight equal sized ponds (1.75 d) situated in the northern sides of the faculty of Fisheries, BAU, were selected and prepared for experiment. The ponds were selected randomly to accommodate the relevant treatments. The experimental layout is shown in Table 35.

Table 35 Experimental layout of Thai Koi and Pangas culture

Treatment	Replications (pond no.)	Pond size (dec)	Stocking density/dec	Total stocked	Stocking size (g)
T_1 (Koi)	R_1 (1) R_2 (2)	0.60 0.60	500 500	300 300	1.20
T_2 (Koi)	R_1 (3) R_2 (4)	0.60 0.60	500 500	300 300	1.20
T_1(Pangas)	R_1 (5) R_2 (6)	0.60 0.60	500 500	300 300	25.10
T_2 (Pangas)	R_1 (7) R_2 (8)	0.60 0.60	500 500	300 300	25.10

Two sets of experiments one with Thai Pangas and the other with Thai Koi were carried out using chemicals and without chemicals (Table 35; Figs. 1and 2). Drugs were applied following fish farmers schedule from pond preparation up to fish culture. Stoking density was maintained at 500/dec. Mega feed was applied at the rate of 5-10% of body weight. Sampling for record of weight and water quality parameters were done once in fortnight (Fig. 3). For health check fish were investigated clinically and samples for histology were taken from skin, muscle, gill, liver and kidney from treated and control ponds of Pangas and Koi once every fortnight. Standard histological procedure was carried out for fixation, dehydration, clearing, infiltration, embedding, sectioning, staining and mounting of slides. Comparisons were made between treated and control treatments of both the Pangas and Koi. Water quality parameters like DO, pH, temperature, alkalinity and nitrite were also recorded fortnightly from all the treatments and replications. Growth parameters like weight gain, percent weight gain, specific growth rate, survival rate, food conversion ratio and production were monitored.

10.7 Results

10.7.1 Categories of chemicals used in aquaculture

Mostly ten different categories of chemicals are found to use in aquaculture activities (Tables 36-46). They are antibiotics, disinfectants, gas removal, oxygen supplier, vitamins and minerals, growth promoter, insect killer, algae killer, predator killer and pH balance.

Table 36 List of antibiotics recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Renamycin	50 mg/kg bw	Renata
02	Aquamysine	1-1.5 kg/ton	Fish Tech
03	Aquamycine	1-2 /kg feed	ACI
04	Renaquine	50 mg/kg bw	Renata
05	Oxy Dox F	1-2 /kg feed	ACI
06	Ascamicyne	250-300 /acre	SKF
07	Amoxivet	25-45 mg/kg bw	Techno
08	Oxysestin	1-2 /kg feed	Novartis
09	Orgamysin	60 /50 kg feed	Organic
10	Doxioxy	1-2 /kg feed	Opsonin
11	Amoxifish	3-5 /kg feed	Fish Tech

Table 37 List of disinfectants recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Polgard plus	500 ml/acre	Fish tech
02	Bactisal	350 ml/acre	First care
03	Virex	100-200 /33 dec	ACI
04	Biogaurd	250 g/33 dec	Sia agro-vet
05	Lenocide	5 ml/dec	Nature care
06	Timsen	1^{st} dose: 80 /33 dec 2^{nd} dose: 50 /33 dec	Eon
07	Emsen	1st dose: 80 /33 dec 2nd dose: 50 /33 dec	Ethical drugs
08	Aqua cleaner plus	1 L/50 dec for 3-5 ft depth	Fish world
09	Formalin	1-3 ppm	Chemical seller
10	Bleaching powder	60 ppm	Chemical seller

Table 38 List of gas removal recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Ammonil	100-200/acre periodically used every 1 month	Novartis
02	Aqua magic	05-08 kg/acre	Fish tech
03	Miracol lime mila	100 /dec	ACME
04	Geolite gold	200-250 /dec (repeat 30-40 days interval)	Fish tech
05	Gas stop	400-500 /acre	Organic
06	Gas check	200 /acre	First care
07	Megageo plus	200/dec	ACI
08	Geo tox	20-25 kg/100 dec (repeat 30-40 days interval)	Novartis
09	Gasonex plus	200-400 /acre	Fish tech

Table 39 List of oxygen supplier recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Oxy gold	250-500 /acre	Fish tech
02	Oxy life	400 /acre/3-6 ft water depth	Square
03	Pure oxy	1-2 kg/1000 m^2	Al Madina
04	Oxymax	250-500 /acre/meter depth	Eon
05	Oxy flow	250-350 /acre	Novartis

Table 40 List of vitamins available in market

Sl.	Trade name	Dose	Source
01	Grow fast	1 ml/3-4 L	Rals
02	Revit C	1/5-7 kg feed	Opsonin
03	Silver mil	1-2 ml/kg feed	Sia agro-vet
04	Ossi-C	4-5 /kg feed for 5-7 days	Fish tech

Table 41 List of growth promoter recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Charger gel	2-4 /kg feed	Fish tech
02	Aqua boost	50 /metric ton	Novartis
03	Bio-grow	100 ml/33 dec	Sia agro-vet

Table 42 List of enzyme recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Biozyme	25-50 /100 kg feed	Fish tech

Table 43 List of insect killer recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Sumithione	5-10 ml/dec/3ft depth	Samco
02	Deletix	25-30 ml/acre for 4 feet water depth	Fish tech

Table 44 List of predator killer recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Rotenone	15 /dec/1 ft depth	Samco

Table 45 list of algae killer recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Seaweed	2-4 liter/acre	Fish Tech

Table 46 List of pH balance recorded from the investigated areas

Sl.	Trade name	Dose	Source
01	Bio-pH	1 ml/L	Biopharma

In pharmacy, around 50 different types of aquaculture drugs and chemicals were recorded. Among those, 15 types are widely used by the farmers as given in Table 47.

Table 47 Mostly used aqua-chemicals in ponds recorded from the investigated areas

Sl.	Trade name	Active ingredient	Purpose of use	Dose	Source
01	Renamycin	Oxytetracycline	Antibiotic	50 mg/kg body weight	Renata
02	Amoxifish	Amoxicillin trihydrate	Antibiotic	3-5 /kg feed for 5-7 days	Fish tech
03	Timsen	n-alkyl dimethyl benzyl ammonium chloride-40%	Killing microbes	1st dose: 80 /33 dec 2nd dose: 50 /33 dec	Eon
04	Aquamysine	Chlorotetracycline	Antibiotic	1-1.5 kg/ton for 5 days	Fish tech
05	Ossi-C	Oxolinic acid, bitaglucan, vit-C	Vitamins	4-5 /kg feed for 5-7 days	Fish tech
06	Aquamycine	Oxytetracycline hydrochloride	Antibiotic	1-2 /feed	ACI
07	Virex	Per oxy monosulfate, sodium hydrochloroisocyanorate	Disinfectant	100-200 /33 dec	ACI
08	Aqua kleen	Tetra decile trimethyl ammonium bromide, benzal conium chloride, amino nitrogen	Disinfectant	0.5-1 L/acre	Square
09	Geolite gold	SiO_2, Al_2O_3, Fe_2O_3, TiO_2, MgO	Gas removal	200-250 /dec (repeat 30-40 days interval)	Fish tech
10	Oxy Dox F	Oxytetracycline hydro-chloride, doxycycline	Antibiotic	1-2 /kg feed	ACI
11	Polgard plus	3-methyl 4 alkyl two chain brominated compound	Disinfectant	500 ml/acre	Fish tech
12	Charger gel	1-3 D glucan, poly-saccharides, betain, bitaglucans	Growth promoter	2-4 /kg feed	Fish tech
13	Seaweed	Elemental copper, inert ingredient	Algae killer	2-4 L/acre	Fish tech
14	Bactisal	Alkyl benzyle dimethyl ammonium chloride	Disinfectant	350 ml/acre	First care
15	Deletix	Deltamethrin	Argulus killer	25-30 ml/acre for 4 feet water depth	Fish tech

10.7.2 Fish production in farmers' pond

Productions of Pangas and Koi in Gouripur and Muktagacha Upazillas were almost double in the chemical treated ponds compared with non treated ponds (Table 48). However, Pangas production in Muktagacha was almost double than Gouripur, where stocking density was also higher (Table 48).

Table 48 Fish production in different areas of Mymensingh region

Area	Species	With chemical/dec	Without chemical/dec	Comments/ Stock Density
Gouripur	Pangas	81 kg	48 kg	Mixed culture
Gouripur	Shing	70 kg	35 kg	200/dec
Muktagacha, Baruri	Pangas	160 kg	120 kg	450/dec
Muktagacha, Baruri	Koi	140 kg	100 kg	2000/dec

10.7.3 Impact of aqua drugs on fish health and diseases

In Mymensingh regions most farmer culture Thai Pangas, Thai Koi, Tilapia and Shing intensively. Productions of Pangas and Koi in Gouripur and Muktagacha Upazillas were almost double in the chemical treated ponds compared with non treated ponds (Table 48). Pangas production in Muktagacha was almost double than Gouripur, where stocking density was also higher (Table 48). In Upazillas like Fulpur, Muktahacha and Fulbaria EUS, dropsy, edwardsiellosis diseases were noticed with Pangas and Tilapia. Shing had 90-100% mortalities within very short period from unknown diseases with no obvious symptoms but swollen abdomen and spots providing 30-00% recovery even after application of drugs. In Fulpur farmers used Renamycin, Ossi-C and Polgard plus and in Fulbaria they used Polgard plus and Bactisol for the treatment of Shing (Table 49). EUS affected Tilapia in Fulpur (20%) and Muktagacha (30%) and farmers used Renamycin, Polgard plus and Ossi C with a result of 95-80% recovery. Edwardsiellosis affected Pangas in Fulpur (80%) and Fulbaria (50%) and farmers used Renamycin, Timsen, Polgard plus and Ossi C in Fulpur and Geolite and Timsen in Fulbaria having 80% recovery in both Upazillas. Dropsy was seen with Tilapia in Fulpur Upazilla (10%) where farmers used Aquamycin and Ossi C as drugs with a result of 95% recovery (Table

49). In Fulpur (50%) and Fulbaria (50%), Zoothamnium and various spots on skin, scales drops in some parts of Koi, where farmers used Renamycin, Aquamycine, Ossi-C and Polgard plus drugs as treatments and achieved 70-80% recovery. *Puntius sarana*, *Labeo rohita*, *Catla catla* and *Cirrhinus mrigala* were also affected by EUS in Muktagacha and the farmers achieved good recovery by applying drugs like Renamycin and Ossi C. The symptoms of diseases, doses of drugs and the affected months were mentioned in the Table 49.

Table 49 Impact of aqua drugs on fish health and disease recovery in the investigated Upazillas

Area	**Species**	**Diseases (Prevalence %)**	**Symptoms**	**Drugs and Doses used**	**Recovery**	**Affected months**
Fulpur	Tilapia	EUS (20%)	Red spot on body surface, lesion on body surface	Renamycin @ 50 mg/kg body weight Polgard plus @ 500 ml/acre Ossi-C @ 3 g/kg feed	95%	August
	Tilapia	Dropsy (10%)	Swelling of abdomen, accumulation of fluid in abdomen	Aquamycine @ 1-2 g/feed Ossi-C @ 3 g/kg feed	95%	August
	Pangas	Edwardsiellosis (80%)	Exophthalmia, red spot on abdomen and dorsal side	Renamycin @ 5g/kg feed Timsen @ 1st dose: 80 g/33 dec 2nd dose: 50 g/33 dec Ossi-C @ 3 g/kg feed Polgard plus @ 5 ml/dec	80%	November-December
	Koi	(50%)	Zoothamnium and red spot on abdomen, scaleless	Renamycin @ 5g/kg feed Ossi-C @ 3 g/kg Polgard plus @ 5 ml/dec	70%	November-December
	Koi	(30-40%)	White spot on tail, gill	Renamycin @ 5g/kg feed Ossi-C @ 3 g/kg feed Polgard plus @ 5 ml/dec	80%	March-April

Area	Species	Diseases (Prevalence %)	Symptoms	Drugs and Doses used	Recovery	Affected months
	Shing	(90%)	No symptoms, sudden death	Virex @ 100-200 g/33 dec Aquamycine @ 1-2 g/feed Renamycin @ 5g/kg feed	30%	November-December
Muktagacha	Shing, Magur	(10%)	No symptoms, sudden death, slightly red line on body surface	Aquakleen @ 0.5-1 liter/acre Gas check 200 g/acre Bleaching powder @ 700 g/32 dec Renamycin @ 5g/kg feed Ossi-C @ 3 g/kg feed Polgard plus @ 5 ml/dec Virex @ 100-200 g/33 dec Aquamycine @ 1-2 g/feed	95%	March-April
	Tilapia, Sarpunti, Rui, Catla, Mrigal	EUS (30%)	Red spot on body surface, lesion on body surface	Renamycin @ 50 mg/kg body weight Ossi-C @ 3 g/kg feed	80%	August
	Pangas	(40%)	Red spot on operculum, spoilage on body surface	Renamycin @ 5g/kg feed Geolite gold @ 200-250 g/dec Aqua kleen @ 0.5-1 L/acre	80%	November-December
	Shing	(40%)	Tail rot	Polgard plus @ 5 ml/dec Aqua kleen @ 0.5-1 L/acre Geolite gold @ 200-250 g/dec	80%	November-December
	Koi	(50%)	White spot on tail, gill	Aquamycine @ 1-2 g/feed Ossi-C @ 3 g/kg feed Polgard plus @ 5 ml/dec	70%	November-December

Area	Species	Diseases (Prevalence %)	Symptoms	Drugs and Doses used	Recovery	Affected months
Fulbaria	Pangas	(40%)	Red sign on eye, fin and heat; Movement on surface	Geolite @ 200-250 g/dec Timsen @ 1st dose: 80 g/33 dec 2nd dose: 50 g/33 dec	80	
	Shing	(100%)	Swollen abdomen with occasional spots	Polgard plus @ 5 ml/dec Bactisol @ 350 ml/acre for 3 ft depth	00%	December-January
	Pangas	Edwardsiellosis (50%)	Speedy movement before death, mouth remains in water surface, Exophthalmia, red spot on abdomen and dorsal side	Renamycin @ 5g/kg feed Ossi-C @ 3 g/kg feed Polgard plus @ 5 ml/dec Geolite gold @ 200-250 g/dec Gasonex plus @ 200-400 g/acre	70%	December-January

10.7.4 Pond experiment at BAU

It was observed that growth of Pangas was significantly higher in the non treated treatment than that of treated treatment at the end of the experiment (Table 50). Whereas, growth of Thai Koi was slightly higher in the treated group than that of non-treated group. However, there were no significant differences of growth between the groups (Table 51).

Table 50 Growth of Pangas in chemical treated and non treated ponds

Sampling No.	Weight (g)			
	T_1R_1	T_1R_2	T_2R_1	T_2R_2
01	25.10±1.56	25.10±1.56	25.10±1.56	25.10±1.56
02	57.00±3.37	57.66±2.58	59.00±2.54	61.00±4.12
03	83.67±4.79	74.33±3.48	75.33±6.06	91.00±6.59
04	109.00±4.81	103.00±6.32	107.33±4.88	129.67±5.95
05	140.67±8.98	126.00±7.64	138.67±6.26	166.67±9.63
06	161.33±4.70	145.67±4.58	161.33±5.04	195.67±17.73
07	170.00±3.94	150.33±4.01	168.67±4.38	204.33±15.60
Average	**160.17**^{a}		**186.50**^{b}	

Table 51 Growth of Thai Koi in chemical treated and non treated ponds

Sampling No.	Weight (g)			
	T_1R_1	T_1R_2	T_2R_1	T_2R_2
01	1.20±0.11	1.20±0.11	1.20±0.11	1.20±0.11
02	3.05±0.08	3.26±0.12	2.78±0.13	2.27±0.27
03	7.20±0.47	7.00±0.48	6.67±0.50	6.67±0.50
04	13.33±0.80	13.33±0.63	15.33±1.98	19.00±2.50
05	27.33±2.17	34.00±4.06	28.33±2.79	25.67±3.30
06	44.67±2.70	36.67±2.11	41.00±2.59	43.67±1.92
07	45.33±2.10	42.33±1.88	44.67±1.86	43.00±2.21
Average	**43.83**^{a}		**43.84**^{a}	

10.7.5 Food conversion ratio and production

Food conversion ratio (FCR) was 1.27 and 1.13 in the treatment T_1 and treatment T_2 in case of Pangas (Table 52). However, in case of Thai Koi, FCR in the Tl was significantly reduced that of T2 (Table 53). Production of Pangas in the non treated ponds was higher which was 7328.16 kg per acre than in the treated ponds, where it was 6400.08 kg per acre. Whereas, production of Koi in the treated ponds was higher (1471.92 kg/acre) than that of non-treated ponds (1296.00 kg/acre). Food conversion and survival of Koi and Pangas are shown in Tables 52, 53, 54 & 55.

Table 52 Food conversion ratio (FCR) of Pangas

Treatments FCR	T_1R_1	T_1R_2	T_2R_1	T_2R_2
Feed (g)	77283.70	63717.37	66194.54	77114.14
Weight (g)	45730.00	36830.85	39637.45	52104.15
FCR	1.69	1.73	1.67	1.48
Mean	**1.71^{a}**		**1.58^{b}**	

Table 53 Feed conversion ratio (FCR) of Koi

Treatments FCR	T_1R_1	T_1R_2	T_2R_1	T_2R_2
Feed (g)	16298.40	15344.63	17503.49	16658.20
Weight (g)	10652.55	10582.50	11078.16	11180.00
FCR	1.53	1.45	1.58	1.49
Mean	**1.49^{a}**		**1.54^{a}**	

Table 54 Survival rate of Pangas

Per pond	Pangas			
	T_1R_1	T_1R_2	T_2R_1	T_2R_2
Stocking	300	300	300	300
Harvesting	269	245	235	255
Survival rate (%)	89.67	81.67	78.33	85.00
Average	**85.67^{a}**		**81.67^{a}**	

Table 55 Survival rate of Koi

Per pond	Koi			
	T_1R_1	T_1R_2	T_2R_1	T_2R_2
Stocking	300	300	300	300
Harvesting	245	260	260	275
Survival rate (%)	81.67	86.67	86.67	91.97
Average	**84.17^{a}**		**89.17^{a}**	

10.7.6 Water quality parameters

Water quality parameters like DO, pH, Temperature, ammonia and nitrite were within the suitable range of fish culture and did not show marked variations among the treated and control ponds in case of both the Thai Pangas and Thai Koi (Table 56).

Table 56 Water quality parameters in the ponds of treated and control treatments

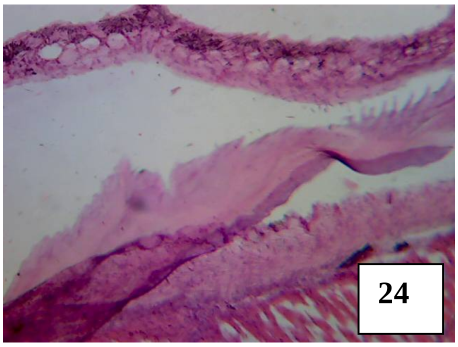
Date	Parameters	Pangas				Koi			
		Treated		Control		Treated		Control	
		T_1R_1	T_1R_2	T_2R_1	T_2R_2	T_1R_1	T_1R_2	T_2R_1	T_2R_2
30.06.11	pH	8.0	7.9	7.9	7.3	7.5	7.2	7.4	7.3
	Nitrite	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
	Ammonia	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	DO	5.5	5.0	5.5	5.5	5.0	6.0	5	5.5
	Temperature	29.0	29.5	30.0	29.9	30.5	30.0	30.0	29.5
22.06.11	pH	7.8	7.9	8.0	8.0	7.8	7.5	8.0	8.0
	Nitrite	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
	Ammonia	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	DO	6	6.5	5.5	6	6	5	5.5	6
	Temperature	30.0	30.5	29.0	29.5	31.0	31.0	29.0	29.5
10.07.11	pH	7.6	7.6	7.7	7.5	7.4	7.6	7.7	7.5
	Nitrite	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
	Ammonia	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	DO	4	4	4	4	4	4	4	4
	Temperature	31.0	30.5	31.0	29.4	30.0	31.0	30.5	29.4
25.07.11	pH	7.6	7.7	7.5	8.0	7.8	7.5	7.8	8.0
	Nitrite	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
	Ammonia	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	DO	6.0	5.0	5.5	5.5	5.5	5.0	6.0	5.5
	Temperature	29.0	31.0	29.5	29.9	30.0	30.5	29.0	29.9
10.08.11	pH	8.5	8.0	7.5	8.0	7.5	7.5	8.0	8.0
	Nitrite	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
	Ammonia	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	DO	4.5	5.5	5.0	6.5	6.0	4.5	5.0	6.5
	Temperature	30.0	29.0	29.0	29.5	31.0	28.5	30.0	29.5

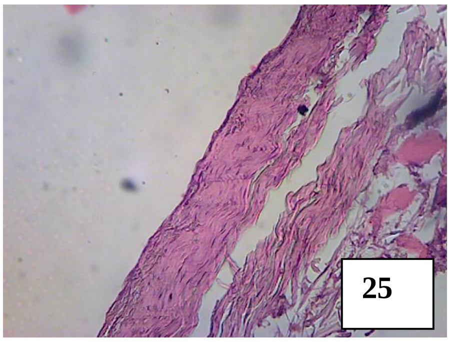
10.7.7 Effects of chemicals on plankton

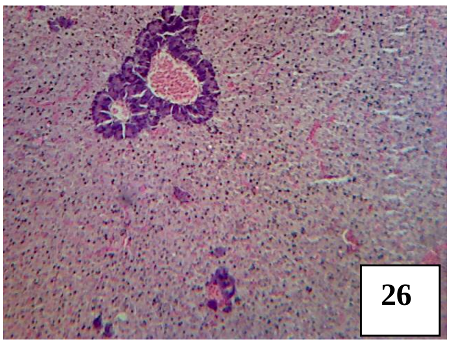
In treatment T_1 and replication R_1, plankton bloom was seen after one month of the start of the experiment. Seaweed (Algicide) was applied at a dose of 20 ml/dec and it was observed that after 7 days of the application of the drug, plankton bloom was disappeared from the pond.

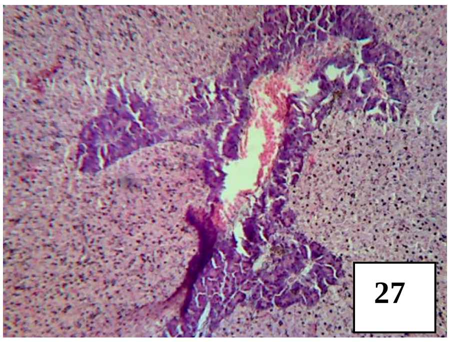
10.7.8 Histopathology for health check

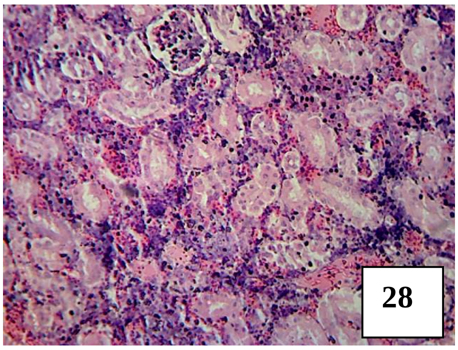
Clinically all the fishes of various regions did not show any remarkable changes. Histopathology in the control ponds, skin-muscle, liver, kidney and gill of fish had almost normal structure (Figs. 24, 26, 28 & 30). However, in the chemical treated ponds, the above mentioned investigated organs of fishes had remarkable pathological changes like necrosis, hemorrhage, vacuum, melanocytes and partial loss of organs (Figs. 25, 27, 29 & 31).

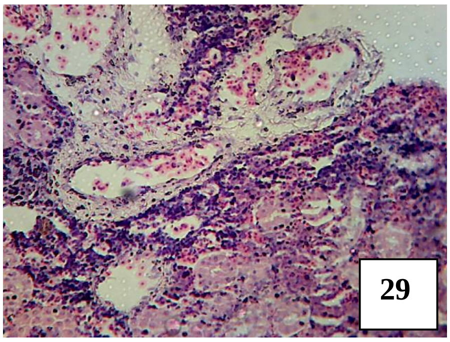

24

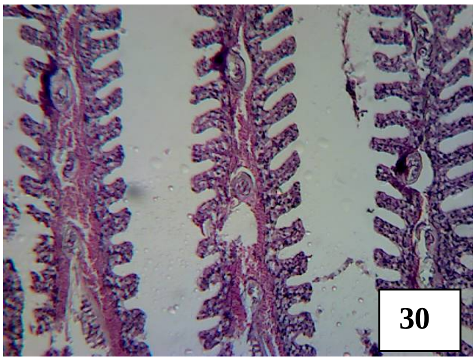

25

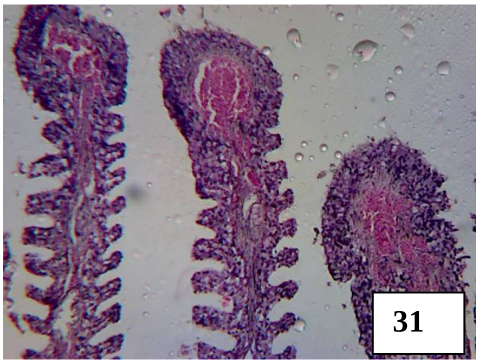

26


27









Figs. 24 and 26 Sections of almost normal skin-muscle and liver of Pangas from Gouripur

Figs. 25 and 27 Sections of affected skin-muscle and liver of Pangas from Gouripur

Figs. 28 and 30 Sections of normal kidney and gill of Pangas from Muktagacha

Figs. 29 and 31 Sections of affected kidney and gill of Pangas from Muktagacha

(All the sections: Haematoxylin and Eosin x 150)

10.7.9 Identified problems

- Farmers use drugs and chemicals without having knowledge on their application.
- In the application of drugs and chemicals withdrawal periods are not maintained.
- Uses of drugs had some adverse effect on fish health.
- Farmers use an increased stocking density of fish which leads to poor health and low production.
- All the drugs of a single company may not suitable for treatment of diseases in a culture system.

10.7.10 Recommendations

- Farmers need knowledge on the use of drugs and chemicals in aquaculture through training.
- Aqua dugs had 70-90% recovery of diseases like EUS, Edwardsiellosis and dropsy.
- It was clearly observed that there are distinct impacts of Aqua drugs on fish diseases, health and production.
- Aqua drugs used farmers' fish ponds yielded almost double production in Muktagacha and Gouripur compared to non-treated fish ponds.
- In BAU experimental ponds, chemical treated and non-treated ponds exhibited almost similar production and lower FCR, in case of Thai Koi. Whereas, in case of Pangas, production was increased in control ponds. So, more research is needed to draw final conclusion.
- Seaweed exhibited positive effect in control of algae in BAU experimental pond.
- Histologically, fish organs of chemical used farmers ponds were affected, whereas, in the control ponds the organs had almost normal structure.
- Although externally, fishes looked normal in the chemical used ponds, some pathological changes were seen in their organs, which show the chemical treated fishes remain in the stressed condition.
- In BAU fish ponds, chemicals used from one company and needs more time to verify with drugs of other companies.
- Present research was conducted in Mymensingh region. Thus other parts of the country and coastal belt need to be investigated.
- Proper withdrawal period should maintain for use of drugs and chemicals.

10.7.11 Conclusion

Some remedial measures should sort out to overcome pathological changes in fish through manipulation of drugs i.e., reduction in number of drugs for single treatment, doses change and use of similar drugs of other companies in order to obtain healthy fish. Aqua drugs and chemicals in aquaculture and fisheries is a vast area of research in

Bangladesh. So, conducting elaborate research is needed to draw a final conclusion on the impact of aqua drugs and chemicals on fish health and productivity.

11. Influence of Chemicals and Drugs on Microbial Flora used Indiscriminately in Aquaculture

11.1 Background and Justification

Microorganisms reside in the water and sediment of aquaculture facilities, as well as in and on the cultured species. They may have positive or negative effects on the outcome of aquaculture operations. Positive microbial activities include elimination of toxic materials such as ammonia, nitrite, and hydrogen sulfide, degradation of unused feed, and nutrition of aquatic organisms such as shrimp, fish production. These and other functions make microorganisms key players in the health and sustainability of aquaculture. Although development of aquaculture in different aspects is notable, microorganisms are among the least known and understood elements in aquaculture yet. Aquaculture of finfish, crustaceans, mollusks, and algal plants is one of the fastest-growing food-producing sectors in the world, having grown at an annual rate of almost 10% from 1984 to 1995 compared with 3% for livestock meat and 1.6% for capture fisheries production Disease outbreaks (EUS in finfish, white spot disease in shrimp) are being increasingly recognized as a significant constraint on aquaculture production and trade, affecting the economic development of the sector in many countries. Conventional approaches, such as the use of chemicals and antimicrobial drugs, have had limited success in the prevention or cure of aquatic disease; a growing concern about the use and, particularly, the abuse of antimicrobial drugs in aquaculture. The massive use of antimicrobials for disease control and boost-up aquaculture production encourages the natural emergence of bacterial resistance which leads ultimate hazard to human health

11.2 Objectives

i. To investigate different chemicals and drugs causing any changes in the normal commensal flora in aquatic habitat and in fish.

ii. To investigate whether the use of these agents influence on post-harvest quality of fish

iii. To check if the use of antibiotics causing the development of antibiotic resistant bacteria

11.3 Methodology

Selected Fish Species for undertaking experiments are:

- Ruhu (*Labeo rohita*)
- Mrigel (*Cirrhinus mrigala*)
- Silver carp *(Hypophthalmichthys molitrix)*
- Common carp (*Cyprinus carpio*)
- Sarputi *(Puntius sarana)*
- Selected Antibiotics:
- Oxytetracyclin (Renamycin)

Selected Chemicals for undertaking experiments are:

- Lime
- Zeolite
- Oxyflow

11.4 Results

Different types of bacteria are found in aquatic habitat (Table 57). Load, concentration and variety of bacteria depend on the quality of water.

Table 57 Common bacterial flora in aquatic habitat

Sl	Bacteria in aquatic habitat
1	*Aeromonas* spp.
2	*Chryseomonas* spp.
3	*E. coli*
4	*Edwardsiella* spp.
5	*Enterococcus* spp.
6	*Flavobacterium* spp.
7	*Listonella* spp.
8	*Mycobacterium* spp.
9	*Mycobacterium* spp.
10	*Pasteurella* spp.
11	*Photobacteriumk* spp.
12	*Pseudomonas* spp.
13	*Salmonella* spp.
14	*Serratia* spp.
15	*Shewanella putrefaciens*
16	*Staphylococcus* spp.
17	*Tenacibaculum* spp.
18	*Vibrio* spp.
19	*Yersinia* spp.

Water quality parameters were more or less similar in antibiotic treated ponds when comparing to control (Table 58). However, microbial load decreased significantly in antibiotic treated ponds as compared to control (Tables 59 & 60). It may be said that water quality parameters of antibiotic treated pond are found suitable for aquaculture

Table 58 Water Quality Parameters of the experimental pond

Parameters		Control Pond	Oxytetracyclin Treated Pond	
			1 week Treatment	Continuous Treatment
Water temperature (^oC)		30.04 ± 1.046	30.41 ± 1.017	30.18 ± 0.541
Transparency (cm)		45.00 ± 7.360	50.65 ± 4.665	38.15 ± 9.797
DO (mg/L)	Morn.	5.66 ± 0.142	5.37 ± 0.210	5.29 ± 0.137
	Even.	7.16 ± 0.148	6.77 ± 0.210	6.89 ± 0.244
pH		7.75 ± 0.165	7.77 ± 0.101	7.69 ± 0.100
Alkalinity		97.38 ± 8.996	98.31 ± 7.028	100.69 ± 8.654
NH_3-N (mg/L)		0.24 ± 0.044	0.23 ± 0.030	0.25 ± 0.020
NO_3-N (mg/L)		0.12 ± 0.010	0.12 ± 0.015	0.14 ± 0.023
NO_2-N (mg/L)		0.024 ± 0.005	0.023 ± 0.004	0.020 ± 0.003

Table 59 Change in microbial load in ponds due to application of antibiotics

Treatments		**Initial Bacterial Load (CFU/g)**	**Final Bacterial Load (CFU/g)**
Control	Without antibiotics	16.6×10^6 to 17.1×10^6	2.3×10^6 to 9.1×10^6
Oxytetracyclin Treated Ponds	5 days Treatment	22.3×10^6 to $23.3\times10^{6*}$	7.75×10^6 to $9.95\times10^{6*}$
	Continuous Treatment	14.4×10^6 to $24.7\times10^{6*}$	2.6×10^6 to $2.6\times10^{6*}$
Lime Treated Ponds	5 days Treatment	19.1×10^6 to 30.1×10^6	2.65×10^6 to 3.54×10^6
	Continuous Treatment	22.6×10^6 to 28.6×10^6	2.9×10^6 to 5.1×10^6
Oxyflow Treated Ponds	2 weeks Treatment	31.6×10^6 to 33.4×10^6	10.1×10^6 to 12.1×10^6
	Continuous Treatment	22.6×10^6 to 24.6×10^6	5.7×10^6 to 6.2×10^6

Table 60 Microbial load in culture ponds at Mymensingh region

Area/Region	Media	Initial Bacterial Load (CFU/g)	Major bacterial flora
Fulpur	Pond water	2.7×10^6 to 1.4×10^6	*Aeromonas* sp., *Shewanella* sp., *Pseudomonas* sp., *Corynebacterium* sp., *Escherichia coli*, and *Clostridium* sp.
	Pond sediment	5.2×10^7 to 17.1×10^7	
Gouripur	Pond water	2.3×10^6 to 3.3×10^6	
	Pond sediment	8.4×10^7 to 22.7×10^7	
Trishal	Pond water	1.1×10^6 to 3.1×10^6	
	Pond sediment	4.7×10^8 to 8.9×10^8	
Bhaluka	Pond water	3.5×10^6 to 5.3×10^6	
	Pond sediment	6.9×10^7 to 4.6×10^7	

11.5 Discussion

Water quality parameters were more or less similar in antibiotic treated ponds when comparing to control. Microbial loads were more or less similar in chemical treated ponds when compared to control ones. Microbial load decreased significantly in antibiotic treated ponds as compared to control. Microbial load in pond water and sediment in different culture ponds of Fulpur, Gouripur, Trishal and Bhaluka varied considerably, suggesting regional variation and poor sanitary condition in the culture ponds of those areas.

12. Determination of Safety Doses of Aqua Drugs in Bangladesh

12.1 Background and Justification

Aquaculture is the farming of aquatic organisms, including finfish and shellfish, by individuals, groups or corporations using interventions (e.g., feed, medications, controlled breeding, and containment) that enhance production. The widespread decline (through over fishing) of many species targeted in capture fisheries continues to drive expansion of the aquaculture industry. The global aquaculture industry is dominated primarily by production facilities located in a few Asian countries: eleven of the top 15 aquaculture-producing countries, accounting for 94% of total global production, are in Asia. China alone accounts for approximately 71% of total global aquaculture production. The magnitude of recent increases in aquaculture production is enormous: annual aquaculture production has more than tripled within the span of 15 years, from 16.8 million tons in 1990 to 52.9 million tons in 2005 (FAO and Fishery Information Data and Statistics Unit, 2005). According to the latest predictions by the United Nations Food and Agriculture Organization (FAO), total annual global fish production (wild and farmed) is expected to increase steadily, from 129 million tons in 2000 to 172 million tons by 2015, with aquaculture accounting for as much as 73% of the total increase. The increase in seafood production through aquaculture provides a good source of high-quality protein and is an important cash crop in many parts of the world. An estimated 56% of the world's population obtains at least 20% of their animal protein intake from finfish and shellfish (FAO 2004).

The intensive aquaculture methods that are practiced in Asia and throughout the world can vary significantly from place to place. However, the majority of aquaculture facilities worldwide rely heavily on the input of formulated feeds and the application of agrochemicals, antibiotics and other inputs, resulting in the presence of many chemical and biological contaminants in aquaculture facilities. This is particularly true in many Asian countries which together produce over 90% of the aquaculture products that are distributed worldwide (to both developing and developed countries. Antibiotics are a group of natural or synthetic compounds that kill bacteria or inhibit their growth and heavy amounts of antibiotics are administered in fish feed for prophylactic (disease prevention) and therapeutic (disease treatment) purposes in aquaculture facilities worldwide (GESAMP 1997; Alderman & Hastings 1998, Graslund & Bengtsson 2001, Holmstrom *et al*. 2003, FAO 2004, Cabello 2006). Despite the widespread use of antibiotics in aquaculture facilities, limited data are available on the specific types and amounts of antibiotics used. The limited usage data that do exist generally originate from developed countries, while the majority of aquaculture production takes place in developing countries where there are limited or no regulatory guidelines in place (Howgate 1998). Even when antibiotic usage data are available in different countries, the same antibiotic products are often marketed under different names and the active ingredients frequently are not listed. Moreover, aquaculture farmers or workers who administer the antibiotics often lack information and/or education regarding the safe and efficient use of these drugs (Graslund *et al*. 2003, Holmstrom *et al*. 2003) potentially resulting in excessive usage that invariably goes unreported. However, there was a substantial lack of quantitative information including the type and total amount of a particular antibiotic used per year on a country-by country basis.

The issue of chemical residues present in seafood, particularly antibiotics, has been thrown into sharp relief by reports of residues of nitrofuran antibacterial being detected in shrimps imported into the EU from Bangladesh, Vietnam and from Thailand. Nitrofuran antibacterials are veterinary drugs whose use in food producing animals and fish is banned in the EU because of health concerns, including a possible increase in cancer risk in humans.

Chloramphenicol has been detected in shrimps imported from Bangladesh, Burma and India. Chloramphenicol, a broad spectrum antibiotic which has latterly been associated with a plastic anemia in humans, has been banned in the EU for use in food producing animals and fish. This is a drug of last resort in human medicine for *Salmonella typhimurium* infections. The EC has responded to these findings by imposing a requirement on member states to monitor imported shrimp from these areas for the specific residues. (Commission 4 GRL_TN_06_2002 Decisions: 2002/249/EC; 2002/250/EC; 2002/251/EC, CEC 2002a, b, c). At the same time, there are relatively few constraints on chemical usage in aquaculture and many antibiotics are widely available. In Bangladesh, a number of chemicals are used in shrimp and finfish farms but their generic names are not known. It is therefore, important to identify safe chemicals and its doses for sustaining aquaculture industry in Bangladesh.

12.2 Literature Review

The identification of antibiotics in imported shrimp has led to extensive coverage of the issue in the international media and, as noted above, led to EC imposition of monitoring. In relation to coverage of this and related issues in the scientific literature, interrogation of the in-house Science Unit literature database and on-line British Library resources generated a general review on classes and types of chemicals used in aquaculture. No quantitative data were included in this review (Gräslund & Bengtsson 2001).

Antibiotic residues have been detected in a small proportion (8-9%) of tiger prawns tested in the UK (Willis *et al.* 1999). Of 204 prawns tested in 1994, one contained detectable oxolinic acid (a quinolone antibacterial), one contained sulphonamides and 16 showed the presence of oxytetracycline. A more recent evaluation using 98 samples from 17 producers spread over 3 countries showed that of these, 23 showed antibiotic activity, 18 contained residues of the antibiotic trimethoprim and 3 of these also contained gentamicin. The agents responsible for the activity in the remaining seven samples were not identified. Chloramphenicol and oxytetracycline were not found. This issue was flagged by the authors as requiring ongoing surveillance. Various other articles in the specialist journals have dealt with the issue of antibiotic resistance (*e.g.* Sorum 1999 & 2000) and the impact of vaccines on reducing drug use is acknowledged for the Scandinavian sector. An extensive descriptive literature exists on the various diseases which can impact diverse aquaculture operations while others focus on the treatment regimes derived for specific problems. While many these papers contain information of interest, they do not address the issue of residues of chemicals present in the marketed products. Some articles in the literature provide quantitative data on antibiotic usage in aquaculture. These have already been referred to in Johnston *et al.* (1998). Such data have primarily been derived from the Scandinavian and North American aquaculture sector. GESAMP (1997) also provide information on the broad classes of chemicals in use in aquaculture, but provide no quantitative data or information on usage patterns.

With respect to the implications of residues in marketed products, while many papers make generic reference to this as established fact and, therefore, of general concern, there are few data in the literature. Even more general searching of internet resources simply retrieved reporting of the chloramphenicol and nitrofuran issue, together with many resources documenting the identity of chemicals in use in the sector. Other resources detail products registered for use in various legislatures.

In relation to detection of bacteria/viruses in marketed product, little if any information exists, although antibiotic resistance in sediment bacteria impacted by aquaculture activities is now well documented. In addition, the focus of the little research carried out to date has been on antibiotic residues. No data were found on other chemicals such as 5 GRL_TN_06_2002 pesticides and antifoulants known to be used in the sector. Finally, no data were found on residues of synthetic chemicals such as dioxins and PCBs transferred via feedstuffs in use in aquaculture beyond information already reported in EC Reports (see: *e.g*. EC 2000), largely from regulatory surveillance programmes.

Overall, the coverage of this issue in the technical literature suggests that, while the potential for problems is high, it remains poorly researched. A similar conclusion was reached by Gräslund & Bengtsson (2001) in relation to shrimp aquaculture:- *"Theoretically, chemicals other than antibiotics that are added to the shrimp ponds, or by-products from the applied substances, that have a bioaccumulation potential, could be found as residues in the shrimps. However, little attention has been paid to the risk of residues other than antibiotics in farmed shrimps, and no data from such investigations have been found"*.

FAO (2005) listed 26 antibiotics from China, India, Phillipines, Japan, Indonesia, Thailand, Vietnam, Bangladesh, Korea, Chile, Norway, USA, Egypt and Thaiwa. Out of 26 anitibiotics, oxytetracycline followed by chloramphenicol and oxolinic acid were the most commonly used antibiotics, while sarafloxacin and sulfadimidine were the least used antibiotics between 1990 and 2007. Of the top 13 aquaculture-producing countries (excluding Egypt and North Korea), 92% used oxytetracycline and 69% used chloramphenicol and oxolinic acid during this time period. Of the 26 antibiotics considered from the FAO list, on average, countries used 7 antibiotics in the aquaculture industry, with Thailand and Japan using the highest number of antibiotics (13 each).

12.3 Objectives

- To collect information on present status of aqua drugs
- To optimize the safety doses of commonly used aqua drugs
- To observe the water quality changes and survival of cultureable species in aqua drug treated ponds
- To optimize the growth performance of some cultureable species after feeding treated with growth promoting drugs
- To monitor the survival and growth performance of fishes treated with aqua drugs

12.4 Results

Study was also conducted to determine the efficacy and performance of three most commonly used Eon Animal Health Products Ltd. which is mainly used for water quality management. Three selected aqua drugs were JV Zeolite, Oxymax and Bio aqua-50.For each drugs three doses as less than recommended, recommended and more than recommended were used to asses the safe and effective dose which gives best result than others. Poor quality water was collected from selected pond by black colored bottles. The water quality parameters that were determined before and after drug treatment were ammonia (mg/L), nitrite (mg/L), dissolved oxygen (mg/L), pH (hydrogen ion concentration), hardness, (mg/L) and alkalinity (mg/L). Recommended dose for JV-Zeolite is 4 mg/L. Collected water was treated by JV Zeolite with three treatments by recommended, less than recommended and more than recommended doses as 4 mg/L, 2 mg/L and 6 mg/L. For Oxymax recommended, less than recommended and more than recommended doses were relatively 0.00012 g/L, 0.00008 g/L and 0.00016 g/L. For Bio aqua-50 recommended, less than recommended and more than recommended doses was relatively 0.00006 ml/L, 0.00005 ml/L and 0.00009 ml/L. The required water quality parameters were monitored with a certain time interval during experiment before and after drug treatment. Safety doses and effectiveness is determined on the basis of desired water quality change. Water quality measurements and sample collection was made between 7.00 and 12.00 noon, on each sampling day. Ammonia, nitrite, dissolved oxygen, pH, alkalinity and hardness were found varying as per hour and dose of drugs in pond. However, it was found that ammonia, nitrite, alkalinity, dissolve oxygen, hardness, and pH ranged from 0.7 to 4 mg/L, 0 to 0.2 mg/L, 150 to 180 mg/L, 3 to 4.5 mg/L, 100 mg/L and 7.3 to 8.2 during the study period after use of drugs. Due to detrimental effect on human health as well as fish health following drugs have been prohibited by Drug Administration of Bangladesh through notification (Table 61).

Table 61 List of drugs prohibited by drug administration through notification

Sl	Trade Name	Generic Name	Chemical Code
1	Oradon Powder	Cloteracycline 5.5 g, Furaltadone 4g	DAR No-271-3424(V)-00
2	Oradon powder	Cloteracycline 55 g, Furaltadone 40g	DAR No-271-3456 (V)-00
3	Furadone Powder	Furaltadone HCI	DAR No-271-3459(V)-01
4	Furaltadone Powder	Furaltadofue HCI 20G	DAR No-271-3428(V)-01
5	Vetfural Powder	Furaltadone (Vet)	DAR No-327-57(V)-83
6	Furidon-Vet Powder	Furazolidone	DAR No-012-359(V)-83
7	Fultad Powder	Furaltadone (Vet)	DAR No-320-30(V)-83
8	Chloramphenicol 10% Inj	Chloramphenicol 10%	DAR No-34-1508 (V)-78
9	Clortetrasone Inj	Prednisolone acitate, Lidocain, Oxytetracycline Chlorhydrate, Chloramphenicol	DAR No- 102-1559(V)-82
10	Chlorocid Inj	Cloramphenicol	DAR No-63-2244(V)-87
11	Neomasticur Inj	Benzypenicillin, Procain, Cloramphenicol	
12	Synavia (Sachet)	Ascorbic Acid, Tetracycline HCI, Furaltadone HCI	DAR No-244-3007(V)-97
13	Metrijet Inj.	Oxytetracycline HCI+Furazolidone	DAR No-244-3008(V)-97
14	Euter Injector Inj.	Procavie Penicillin G, Diphenylsulphone, Sulphamilamide, Chlorampenicol, Nitrofurazone, Chloramphemicol, Nitrofurazone, Sulphapyridune, Dihydiostreeptomyein Sulphate	DAR No- 196-2207(V)-86
15	Furaltadone 30% Powder	Furaltadone HCI 30G	DAR No- 204-2248(V)-89
16	Neo-uerotab Inj.	Neomycin 100 mg, Nitrofurazone	DAR;No-204-2778(V)-89
17	Leukomycin Sol.	Chloramphenicol 20%	DAR No-61-1237(V)-78
18	Tardomyocel Ointment	Chloramphenicol, Benzypenicillin K, Benzathin Penicillin	DAR No-61-1264(V)-78
19	Lenkomycin 20% Sol	Chloramphenicol	DAR No-61-1264(V)-78
20	Urozol Powder	Furazolidone	DAR No-011-237(V)-83
21	Furasol	Furaltadone	DAR No-308-64(V)-83
22	Furoxin (Vet)	Furaltadone	DAR No-308-57(V)-83
23	Furadon Powder	Furaltadone	DAR No-304-237(V)-83

Sl	Trade Name	Generic Name	Chemical Code
24	Nitrovet Bolus	Nitrofurazone, Urea, Metronidazole	DAR No-304-250(V)-83
25	Chlorsone Inj.	Prednisolone Acctate, Oxytetracycline Chlorhydrate, Chloramphenicol	
26	Furaltadone (Vet) Powder	Furaltadone	DAR No-175-117(V)-83
27	Chloramphenicol (Vet) Powder	Chloramphenicol	DAR No-175-118(V)-83
28	Fural 50% (Vet) Powder	Furaltadone	DAR No-175-138(V)-83
29	Razovet	Furazolidone	DAR No-025- 463(V)-83
30	Metramid bolus	Furazolidone	DAR No-025- 465(V)-83
31	Metrodon Bolus (Vet)	Metronidazole, urazolidone, Loperamide Hydrochloride	DAR No-050-215 (V)-83
32	Zolivet	Furazolidone	DAR No-267-108 (V)-83
33	Furazole Premix	Furuzolidone	DAR No-083-60 (V)-83
34	Furazole Suspension	Furazolidone	DAR No-083-107 (V)-83
35	Furavet WSP	Furazolidone	DAR No-036-256 (V)-83
36	Coccizol Powder	Furazolidone	DAR No-158-050 (V)-83
37	Fural 30 (Powder)	Furaltadone	DAR No-276-33 (V)-83
38	Utepesary Pressary	Nitrofurazone, Neomycin Sulphate	DAR No-276-41 (V)-83
39	Nefidon 200 (Powder)	Furazolidone	DAR No-002- 303 (V)-83

Being a chemical compound, drugs have positive effect on human health as well as on fish. Considering the fact, following drugs have been approved by the EU/USFDA for treating fishes and other aquatic animals (Table 62).

Table 62 EU/USFDA approved drugs for aquaculture and their doses

Sl	Drug Name	Dose
1	Trimethoprim	50ug/kg
2	Amxiocyllin	50ug/kg
3	Ampicilin	50ug/kg
4	Benzylpenicillin	50ug/kg
5	Coxacilin	50ug/kg
6	Decoxacilin	300ug/kg
7	Oxacilin	30ug/kg
8	Flumequine	600ug/kg
9	Saraffloxacin	30ug/kg
10	Chlorotetracycline	100ug/kg muscle, 300ug/kg/liver, 600ug/kg kidney
11	Oxytetracycline	100ug/kg muscle, 300ug/kg/liver, 600ug/kg kidney
12	Tetracycline	100ug/kg muscle, 300ug/kg/liver, 600ug/kg kidney

A variety of chemicals and drugs having different trade and generic names are used in fish culture during pond preparation (Table 63). Water quality management is crucial in fish culture. Different types of chemicals and drugs are used to maintain water quality during fish culture.

Table 63 Chemicals used for pond preparation and water quality management

Trade name	Active ingredients	Doses	Manufacturer
Geotox JV Zeolite	SiO_2, Al_2O_3, Fe_2O_3 CaO, MgO, Na_2O	For 3-6 ft water 20-25kg/100 dec 3.5-7 kg/33 dec, every 15 days	Novartis pharmaceuticals
Green zeolite	SiO_2, Al_2O_3, Fe_2O_3 CaO, MgO, Na_2O	20-25 kg/100 dec (pond preparation), 10-15 kg/100 (during culture	Organic Pharmacuticals
Zeolite	SiO_2, Al_2O_3, Fe_2O_3 CaO, MgO, Na_2O	20-30 kg/acre	Syngenta
Zeocare	SiO_2, Al_2O_3, Fe_2O_3 CaO, MgO, Na_2O	200 g/acre	Nature care
Lime	CaO, $Ca(OH)_2$	Spread 6-10 ppm	Chemical seller
Mega zeo	SiO_2, Al_2O_3, Fe_2O_3 CaO, MgO, Na_2O ,Mn	For 3-6 ft water 20-25 kg/100 dec	ACI animal health
Super Zeolite	Sio_2, Al_2O_3, Fe_2O_3, CaO,MgO, LoI, K_2O	20-30 kg/acre	Avon animal health
Bio Aqua	Extract of Uka cidizera tree	2 ml/100 dec	Eon animal health products
Gastrap	Lactic acid bacillus, *Bacillus subtilis*, amilase, Cellulose, Lipase	200 g/acre	Square
Aquanone	Rotenone	5-7 kg/acre	Square
Zeo- fresh	SiO_2, Al_2O_3, Fe_2O_3, CaO,MgO, LoI, K_2O	20-24 kg/acre	Square
Zeo prime	Sio_2, Al_2O_3, Fe_2O_3, CaO,MgO, LoI, K_2O	20-24 kg/acre	SK+F

Disinfection is one of the processes of pond preparation for fish culture. After every culture cycle, disfection is very important. Different chemicals and drugs are used to disinfect ponds (Table 64). Disinfectant is also used during fish culture as well when water quality become deteriorates.

Table 64 Chemicals used as disinfectant in ponds

Trade name	Active ingredients	Doses form	Manufacturer
EDTA Bleaching	Sodium thiosulphate Chlorine	0.1-1 ppm 60 ppm	Chemical seller Chemical seller
Timsen	n-a alkyl dimethyl benzyl amonium chloride+stabilized urea	20-80g/33 dec	Eon animal health products
Water clear	Sodium thiosulphate	2-3 L/100 dec	Organic Pharmaceuticals
Omicide	Benzil ammonim chloride+urea	200 ml/acre	Lion Overseas
Formalin	38% Formaldehyde	1-3 ppm	Chemical seller
BKC	Benzal Konium chloride	Spread with water 0.5 ppm	Chemical seller
Efinol	Efinol	5-8 g/1000 L water	Eon animal health products
Aquakleen	Tetradesile trimethyle amonium bromide, Aminonitrogen	24 kg/acre	Square
Bio Aqua		2 ml/100 dec	Eon animal health products

Dissolved Oxygen (DO) is crucial for all aquatic life. Due to low DO, fish come to the surface of water and try to engulf oxygen from air. Fish hang from the surface and die due to critically loe DO. A variety of chemicals/ products are available in local market those release oxygen, when applied in ponds (Table 65). Such chemicals/products increase DO instantly in pond water but concentration of DO decreases with passing of time.

Table 65 Chemicals/Products used for oxygen supply in ponds

Trade name	Active ingredients	Doses form	Manufacturer
Oxyflow Oxymax	H_2O_2 10% H_2O_2 10%	250-350 g/acre 250-500 g/acre	Novartis Pharmaceuticals Eon animal health products
Bio-ox	Sodium carbonate, H_2O_2 10%	2.5-5 g/acre	ACI animal health products
Oxyplus	Na_2 O_2 $^{+}AlOHNa_2O_2$ 90%	500 g/acre	Navana animal health
Oxygen plus	O_2 Promoter	250-500 g/acre	Avon animal health
Oxymore	Sodium carbonate peroxyhydrat	250-500 g/acre	SK+F Bangladesh Ltd
Oxygrow	O_2 Promoter	500 g/acre	Century Agro Ltd
Oxylife	Oxygen precursors	400 g/acre	Square
Best oxygen	Sodium percarbonate	200-250 g/acre	Univet Ltd
Oxygold	Sodium percarbonate	200-250 g/acre	Fishtech Ltd
O_2 Marine, Quik Oxygen	Peroxide Oxygen	200-300 g/acre	Organic Pharmaceuticals Ltd

Disease is a critical factor for fish culture in Bangladesh. With the intensification of aquaculture, fish are suffering from different types diseases. Disease out-break in fish pond is a yearly phenomenon in many parts of the country especially during winter season. During winter season, water quality of most ponds become deteriorates due to low depth of water and application of feed. Different types of chemicals and drugs are applied to treat fish (Table 66). In many cases, farmer applies more than one chemicals and drugs to treating fish.

Table 66 Chemicals used for treatment of diseased fish

Trade name	Active ingredients	Doses form	Manufacturer
Potash Lime	$KMnO_4$ CaO, $Ca(OH)_2$	3-5 g/dec 0.5-1.0 kg/dec	Chemical seller Chemical seller
Formalin	40% Formaldehyde	0.1-0.2 ppm	Chemical seller
Salt	$NaCl$	0.25-1.0 kg/dec	Chemical seller
Methylene blue	$C_{10}H_{18} ClN_3 S_x H_2 O$		Chemical seller
Malachite green	$C_2 H_2 O_4$		Chemical seller
Malthion	Malthion	500 g/acre	Century Agro Ltd
Cevit- Aqua	L-Ascorbic acid	1-3 g/kg feed	Square
Best oxygen	Sodium percarbonate	200-250 g/acre	Univet Ltd
Oxygold	Sodium percarbonate	200-250 g/acre	Fishtech Ltd
Registrol	Betain, Calcium, P, Vit-C	5-10 ml/kg feed	Square

Antibiotice are applied to treat diseased fishes in ponds. Both oral administration and injection are practiced in fishes. Generally, required antibiotic is mixed with feed and supplied in pond. In case of large fishes, individual fish can be treated by injecting antibiotics. No antibiotic is especially prepared for treating fish rather it is for other animals. A variety of antibiotics is available in local market and is used for fish treatment (Table 67).

Table 67 Antibiotics used for treatment of diseased fish

Trade name	Active ingredients	Doses form	Manufacturer
Oxysentin 20%	Oxytetracline HCl BP	100-200 g/100 kg feed, 5-7 days	Novartis
Chlorsteclin	Chlorsteclin	200-300 g/100 kg feed	Novartis
Ranamox	Amoxicillin Trihydrate	28-40 g/100 bd of fish	Renata
Renamycin	Oxytetracycline	28-42 g/100 kg feed	Renata
Orgamycin 15 %	Oxytracycline HCl	60 gm/100 kg feed 10days	Organic
Orgacycline-15%	Chlorotetracycline	200-300 g/100 kg feed	Organic
Bactitab	Oxytetracycline 20%	50 g/kg body weight	ACI
Sulfatrim	Sulphadiazine & Trimethoprim	50 g/kg body weight	Square
Oxin WS	Oxytetracycline 20%	50 mg/kg body weight	Square
Cotrim-Vet	Sulphamethoxazole + Trimethoprime	0.50mg/kg body weight	Square