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Research Article

Use of aquadrugs, chemicals and biological products in freshwater aquaculture systems of West Bengal, India

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ABSTRACT

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In Indian aquaculture, the information on the usage of aquadrugs is very little. The present survey focused on the aquadrugs, chemicals and biological products used in 676 freshwater farms from 15 districts of West Bengal between 2018 and 2019. The aquadrugs usage/ton fish production was calculated for each product. A total of 109 different products such as soil and water treatment compounds, disinfectants, piscicide, inorganic fertilizers, antimicrobials, antibiotics, pesticides, insecticides, feed additives, probiotics and herbal products were documented. The farmers of the South 24 Parganas and West Midnapur districts used the maximum (n=48) and least (n=7) number of products, respectively. Lime was the most common product used by the farmers (87.28%) amounting to 48.70±46.62 kg/ton fish production. The input in terms of quantity was dominated by mahua oil cake (233.19±189.87 kg/ton fish production). The antibiotic application was the least in West Bengal aquaculture, i.e., 0.35±0.23 L/ton fish production and 4.59±2.13 mg/ton fish production. The products of pesticides and insecticides were dominated by nuvan and formalin (17.62±9.33 mL/ton fish production) and ivermectin (1.33±1.29 g/ton fish production). The survey results indicated that the quantity of aquadrugs used in West Bengal was lower than those recorded in many other major fish producing countries. Nevertheless, the majority of the farmers lack knowledge on the appropriate dosage, mode of application and the impacts of aquadrugs. This study, therefore, calls for imparting appropriate knowledge on the use of authorized drugs, environmental impacts, standards on drug residues and consumer safety to the fish farmers

INTRODUCTION

Aquaculture has become the major source of global food fish production with the continued decline in fisheries resources from the wild. Globally, China has remained a major fish producer, accounting for 35% of global fish production in 2018. Freshwater aquaculture production is dominated by the culture of several species of carp, tilapia, and catfish in the intensified farms (FAO, 2020). The aquaculture production sector globally has rapidly expanded and intensified to sustain the increase in population demand for consumption and economic growth. According to FAO (2020), the average annual growth rate of aquaculture is expected to slow down from 4.6% in 2007-2018 to 2.3% in 2019-2030 due to intensified aquaculture production practices and increasing outbreaks of diseases. Aquaculture farmers have relied on a wide variety of synthetic and natural chemical and biological treatments to prevent or treat disease outbreaks, enhance the health status of fish and improve the environmental conditions of the aquaculture production systems. These include soil and water treatment, disinfectants, insecticides, antibiotics, fertilizers and other

compounds (Rico *et al*., 2013; Lulijwa *et al*., 2020). Globally, the fish farmers still rely on aggressive use of chemotherapeutics to combat bacterial infections and infestations of ectoparasites as well as disinfectants to manage the spread of diseases. The products that are used for the treatment are mostly regulated in all jurisdictions and can be used under prescription from a licensed veterinarian. A prolonged withdrawal period of the aqua-drugs and medicines is important because of persistent residues (Limbu *et al*., 2020). A particular concern is the potential usage of chemicals and aquadrugs in aquaculture due to poor management may affect the non-target organisms and may trigger antibiotic resistance in microbes (Watts *et al*., 2017).

At present, India contributes about 6.30% to global fish production and 5% to global trade (Singh and Singh, 2018). In India, the state of West Bengal is ecologically well endowed for inland fisheries with 16.19 lakh tonnes of fish production in 2019-20 (DOF, 2020). It is the second-largest fish producing state after Andhra Pradesh (DOF, 2020). In West Bengal, the average productivity in inland ponds under the Fish Farmers Development Agency has been increased from 600-800 kg/ha/year in the year 1981-82 to 4000-4850 kg/ha/year in 2017-18 (Anon, 2019). West Bengal is practising composite fish farming that comes across an incursion of infectious pathogens that leads to the occurrence of disease outbreaks (Abraham *et al*., 2010). Several aquadrugs, chemicals and biological products are used with the increase in aquaculture practices to control the production loss (Mishra *et al*., 2017). Though the West Bengal fish farmers use several aquadrugs in various stages of fish production, there is no systemic documentation of the number of aquadrugs, chemicals and biological products used and their quantities. The present study aimed to assess the freshwater aquaculture fish production in West Bengal and to evaluate the usage of aquadrugs such as disinfectants, antimicrobials, antibiotics, pesticides, feed additives, and probiotics quantitatively per ton fish production.

MATERIALS AND METHODS

Overview and scope

The data on freshwater aquacultured fish production and application of aquadrugs in fish farms located in 15 districts, viz., South 24 Parganas, North 24 Parganas, Howrah, Hooghly, East Midnapur, West Midnapur, East Bardhaman, West Bardhaman, Bankura, Birbhum, Nadia, Purulia, Murshidabad, Malda and South Dinajpur of West Bengal, India were collected (Fig. 1). These districts play a vital role in supplying the food fish to the entire state. A total of 676 freshwater aquaculture farms were surveyed to collect the required information during the survey period of 12 months between 2018 and 2019.

Structured interviews

During the study period, the information on the aquacultured fish production and application of aquadrugs, chemicals and biological products in the farms were collected by a stratified random sampling technique using key participatory rural appraisal (PRA) tools. The basic details on fish farmers were collected initially from the Fishery Extension Officers (FEO) of the Department of Fisheries, Government of West Bengal in each district. The primary data on the application of aquadrugs such as soil and water treatment compounds, disinfectants, antimicrobials, antibiotics, pesticides, feed additives, immunomodulators, probiotics, etc in the farms were collected through on-field and passive surveys through personal interviews. Interviews were also carried out with aquadrugs suppliers and retailers in retail shops located close to the farm areas. Information was collected through a structured questionnaire developed by the ICAR-Central Institute of Brackishwater Aquaculture, Chennai under the All India Network Project on Fish Health. The structured questionnaire comprised of (i) basic information on the respondents with their farming experience, (ii) pond and farm characteristics and management practices, (iii) cultured species stocking, biomass, disease occurrence and actual or expected production, (iv) prophylactic and therapeutic use of aquadrugs and chemicals with quantity and frequency of application and (v) capacity to manage health crisis in aquaculture.

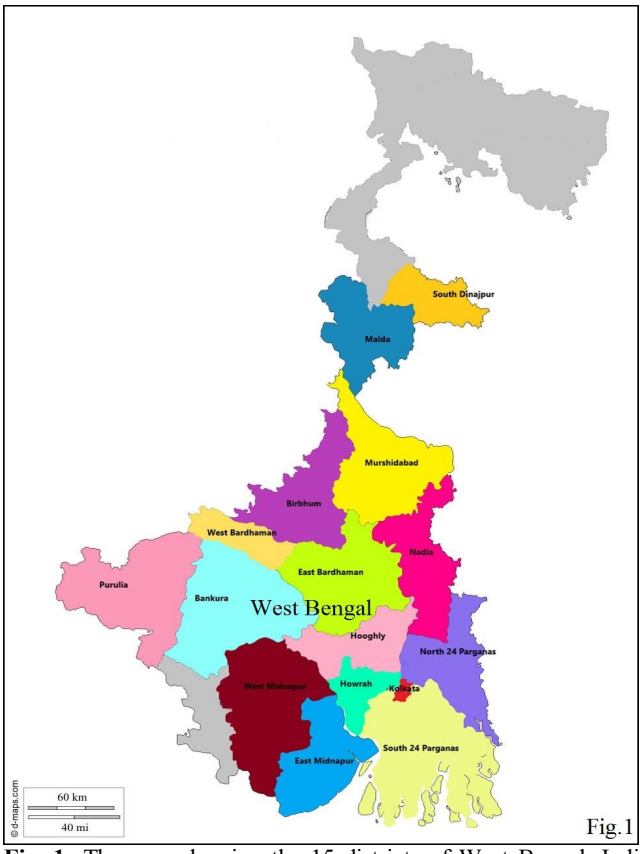

Fig. 1: The map showing the 15 districts of West Bengal, India surveyed on the use of aquadrugs in freshwater aquaculture

Collection of data and classification of aquadrugs

The aquadrugs, chemicals and biological products were classified into ten major categories, viz., (i) soil and water treatment compounds (ii) disinfectants (iii) piscicides (iv) inorganic fertilizers (v) antimicrobials (vi) antibiotics (vii) pesticides and insecticides, (viii) feed additives (ix) probiotics and (x) herbal products. The main active ingredient(s) in each of the reported products was recorded and/or identified by searching the reported products name in the published literature or cross-checking with surveyed reports. Reported dosages of antibiotic, disinfectant and

pesticide compounds were compared with recommended dosages. As farmers typically report dosages in the mass of formulated product/unit area, the reported dosages were recalculated into standard dosage units like mg/L for compounds applied directly to water, or mg/kg biomass of cultured organisms or g/kg feed for compounds applied mixed with feed and compared with the recommended or approved dosage provided by the national and international agencies such as Coastal Aquaculture Authority (CAA), United States Food and Drug Administration (USFDA), etc.

Calculation of aquadrugs usage pattern

Based on the data from the respective district's respondent farmers, the number of aquadrugs used and the total usage of each aquadrug in each district was calculated. The aquacultured fish production in the individual farm of the respondent farmer in each district was collected and converted uniformly to production in kg/ha. The average production of aquacultured fish/ha in each district was calculated from the production data of the respective district's respondent farmers. The aquadrugs usage/ton aquacultured fish production was calculated for each product from the estimated average production data. This was done by calculating the total quantity of each product used in farms relative to mean fish production in each district as given below.

Aquadrugs usage/ton fish production = [Total quantity of each product used in the surveyed farms / average fish production in the surveyed farms] x 1000

RESULTS AND DISCUSSION

Aquadrugs usage in fish farms

A total of 109 numbers of aquadrugs, chemicals and biological products were documented during the study period in 676 surveyed fish farms of West Bengal. The South 24 Parganas district farmers used the maximum number of products (n=48) particularly during the pre-and post-stocking period of culture. The farmers of West Midnapur were the least users of aquadrugs (n=7) (Fig. 2). These products were categorized as soil and water treatment compounds (n=13), disinfectants (n=14), piscicide (n=1), inorganic fertilizers (n=7), antimicrobials (n=23), antibiotics (n=11), pesticides and insecticides (n=12), feed additives (n=17), probiotics (n=8) and herbal products (n=3) (Tables 1-4).

Table 1: Soil and water treatment compounds and disinfectants used in freshwater aquaculture farms of West Bengal

Trade name	Active ingredients	Dose and dosage	Indications	Approval status
Lime	Calcium oxide (CaO), Calcium hydroxide ($Ca(OH)_2$)	Pond preparation: 100-8000 kg/ha Rearing period: 10-500 kg/ha	Maintains healthy pH of pond water, stable planktonic mass, reduce turbidity	NA
Aqualite	Zeolite	180-350kg/ha	Removes hydrogen sulphide, carbon dioxide and ammonia	CAA/APR19/DIS/03404
Dolomite	Calcium magnesium carbonate ($CaMg(CO_3)_2$)	100-300kg/ha	Maintains pH level, increases alkalinity	NA
Toximer	Hydrated sodium calcium alumino-silicates, Mannan oligosaccharides, organic acids, activated charcoal, choline chloride and digestive enzymes	Water: 5 kg/ha	Removes ammonia	CAA/F16/FA/00027
Prolite	Hydrated sodium calcium alumino- silicates, probiotic: *Bacillus subtilis*, *Bacillus licheniformis, Nitrosomonas, Nitrobacter, Aerobactor.*	Pond water: 25 kg/10,000 m^2	Removes the toxicity of ammonia, reduces the turbidity, regulates pond pH	CAA/M16/FA/00460
Aquafresh	Extract of *Yucca schidigera* and aloe vera fortified with special selective species of beneficial bacteria and digestive enzymes	Pond water: 1 kg/10,000 m^2	Eliminates ammonia, Stress reliever, improves feed intake, reduces the pathogenic microbial load.	CAA/MAR 18/FA/01502
Yuccus	*Yucca schidigera*, aloevera, and *Bacillus* spp.	500 g/ha	Eliminates toxic/ pungent odours, reduce harmful bacteria loads	CAA/JAN19/FA/03060
Oxymore plus	Sodium carbonate peroxyhydrate, natural oxygen releasers with concentrated zeolites	Pond preparation: 1-2 kg/ha;	Improves dissolved oxygen levels, prevents mass mortalities, helps to reduce COD and BOD	CAA/APR19/FA/03471
Toxifree	Mannan oligosaccharides organic acids, activated charcoal, choline chloride, digestive enzymes and sodium-calcium aluminosilicates	Water: 5 kg/ha	Adsorbs toxic gases	NA
Amonil	Yucca plant extract	200-250 g/1000 m^2	Removes ammonia, controls water pH	CAA/OCT18/FA/02760
Cure-in	Calcium carbonate, copper sulphate, manganese sulphate, potassium permanganate, magnesium sulphate and essential trace minerals	9.39 kg/ha	Improves the water quality	CAA/JY17/FA/00858
Addoxy	Tetraacetyl ethylene diamine and sodium perborate	500g/ha	Helps to remove the toxic gases	CAA/F16/CHEM/00045
Aqua powder	Active silicon and aluminum	10- 15 kg/ha	Purifies water, improves dissolved oxygen, absorbs ammonia, hydrogen sulphide, carbon dioxide and other toxic gases	CAA/APR19/FA/03376
Gasix	*Yucca schidigera*	200 g/ha	Absorbs toxic gases, reduces turbidity, increases dissolved oxygen levels	CAA/SEP18/PRO/02743 G
Zeodon	Granular zeolite	25-50 kg/ha	Adsorbs toxic gases like ammonia, hydrogen sulphide, carbon monoxide	CAA/MAR 18/FA/01481

Quickphos	Aluminum phosphide (AIP)	0.142 g (1 tablet)/ ha	Rodenticide, insecticide and fumigant (Highly toxic inorganic compound)	NA
Bleaching powder	Calcium hypochlorite ($Ca(ClO)_2$)	3.65g/ha	Disinfectant during pond preparation	NA
Aquamagic	*Azotobacter chorcoccum, Bacillus subtilis* and *Candida utilis*	5 kg/10L (mixed with 100g sugar+250g rice bran)	Disinfectant or toxic gas reducer	CAA/AUG18/CHEM/02374
Sanitiser aqua	BKC-80% and iodine-20%	40 mL/ha	Controls ulcer, tail rot-fin rot, gills infection, dropsy, black spot in shrimp, cotton wool disease, *Saprolegnia* and mass mortality	NA
TH4 plus	Didecyl-dimethyl ammonium chloride, dioctyl-dimethyl dimethyl ammonium chloride and octyl decyl dimethyl ammonium chloride.	140 mL/ha of pond water and broadcast uniformly all over the pond	Bactericide and fungicide. Controls the microbial growth	NA
Blesson	Alkyl dimethyl benzyl ammonium chloride-50%; Stabilizer-6%	4-5 L/ha (1.5 m water depth)	Anti-bacterial, anti-fungal and anti-protozoal	CAA/F16/DIS/00042
Stress free	Natural bacteria	190 mL/L of water for 7 days	Stress reliever	CAA/M16/FA/00354
Iodo – 20	Alkylarylpolyoxyethelene iodine complex:20%	300 mL/ha	Anti-bacterial, anti-fungal, anti- protozoan	FDA low regulatory
Aquakleen	Tetradecylaminonitrogen	59.25 kg/ha	Disinfectant	CAA/MAR 18/DIS/01340
Virkill plus	An active synergistic blend of organic acids, surfactants and high and low molecular weight biocides	Regular use once in 3 weeks: 200 g/ha; During serious outbreaks: 0.404 kg/ha	Disinfectant	CAA/AUG18/DIS/02223
Potash	Potassium permanganate ($KMnO_4$)	1-10 mg/L for 1 hour	Controls external protozoan, metazoan parasites and bacterial and fungal infections	Unapproved by FDA-CVM and IFAS
Betadine	Povidone-iodine	Egg surface-:100 mg/L	Disinfectant during and after water hardening	FDA low regulatory drug, FDA 2011
Cleaner	Cypermethrin	10 mL/ha	Disinfectant	Approved in the United Kingdom, EC regulation 2009

NA: Not available; CAA: Coastal Aquaculture Authority, India; FDA: Food and Drug Administration; CVM: Centre for Veterinary Medicine; IFAS: Institute of Food and Agricultural Sciences, Florida; EC: European Commission; COD: Chemical Oxygen Demand; BOD: Biological Oxygen Demand.

Table 2: Piscicides and insecticides used in freshwater aquaculture farms of West Bengal

Trade name	Active ingredients	Dose and dosage	Indications	Approval status
Mahua oil cake	Saponin	200 mg/L	Poisonous towards lower forms of life and fish	NA
Hitek powder	Ivermectin	200-250 mg/ ton feed	Pesticide	No regulatory approval by CVMP, EPA
Salt	Sodium chloride (NaCl)	0.5-1.0% indefinitely	Anti-parasitic, used as an osmoregulatory and to relieve stress.	FDA low regulatory drug, FDA 2011
Copper (II) sulfate pentahydrate pure	Copper sulfate ($CuSO_4$)	40-50 mg/L for 30 min dip	Removes marine fish ectoparasites	Approved by EPA
Paraclean	Biologically derived macrocyclic lactones with herbal extract, inert carrier and adjuvant	100 g/ton fish biomass for 4-5 consecutive days	Effective against endo and ectoparasites of culture fish including gill parasites and gill flukes	NA
Nuvan	Dichlorvus tech. 83.0%, xylene 8.0%, epichlorohydrin 1.0%, emulsifier 7.0%, triethanonline 0.9%	80 mL/ha	Insecticide	Unapproved by EPA, 1987
Formalin-F™	Formalin	Finfish eggs:1000-2000 µl/L for 15 min; Finfish: 250µl/L for 1hr	Controls Saprolegniaceae	USFDA approved; CAA/JAN19/DIS/03243
Fish ick cure	Malachite green	95 mL/L	Controls ich disease or white spot disease	CAA/JAN19/DIS/03272
Acriflavine	Acriflavine (salts) – euflavine and proflavine	Eggs: 100 ppm dip for 5 sec; catfish: 5ppm for prolonged treatment	Controls external protozoan	CAA/JAN19/CHEM/03273, Unapproved by FDA-CVM and IFAS
Methylene blue	Dimethylaminophenazathionium chloride	2 mg/L prolonged bath	Controls bacterial gill disease and fin rot	Unapproved by FDA-CVM and IFAS

NA: Not available; CAA: Coastal Aquaculture Authority, India; USFDA: The United States Food and Drug Administration; EPA: Environmental Protection Agency; CVMP: Committee for Veterinary Medicinal Products; FDA: Food and Drug Administration; CVM: Centre for Veterinary Medicine; IFAS: Institute of Food and Agricultural Sciences, Florida; EC: European Commission

Table 3: Feed additives, probiotics and herbal compounds used in freshwater aquaculture farms of West Bengal

Trade name	Active ingredients	Dose and dosage	Indications	Approval status
Agrimin	Cobalt, copper, iron, magnesium, manganese, potassium, sodium, sulphur, zinc, DL-methionine	NA	Maintains stable health, stress-free	CAA/F16/FA/00002
Nutriva F 15	Vitamins: A, C, E, D3, B1, B2, B6, B12, niacin, pantothenic acid, folic acid	Shrimp: 5-8 g/kg feed regularly	Stress-free, growth promoter	CAA/AUG18/FAU/02403
Growth plus	Protein hydrolysate, multivitamins, essential minerals, digestive enzymes, probiotics	5-10 g/kg of feed continuously or at least 7 days for 30 days	Supplements more nutrition to fasten the growth.	CAA/AUG18/FA/02355
Fishzyme	Alpha – glucanase: xylanase: pectinase, cellulose, acid protease, mannanase, glucoamylase, amylase	Regular use: 500 g/ton feed, Less nutritional value feed-:1000 g/ton feed.	Improves feed digestion, increases the nutritional value of feed and daily weight gain of fish	NA
Carbozyme	Alcohol, fungal diastase, hydrolyzed casein, papain	250 g/ ton feed	Stimulates appetite	CAA/JAN19/FA/03369
Aquamin	Essential minerals, vitamins, amino acids and antioxidant	100 g/10kg feed	Acts as a growth promoter	CAA/APR19/FA/03478
Mineral mix	Cobalt, copper, iodine, iron, magnesium, manganese, potassium, sodium, sulphur, zinc, DL-methionine, L-lysine monohydrochloride, calcium, phosphorus, base	1-2 kg/ton fish	Improves pond water nutrition, promotes growth	CAA/M16/FA/00516
Aquazyme	Multi enzyme feed additive with vitamins, minerals and amino acids; Vitamins: A, D3, E, calcium, phosphorus, copper, cobalt, magnesium, zinc, iodine, iron, L-lysine, DL-methionine, sulphur, potassium, sodium, selenium, yeast	10-20 g/1000 L for 7 days	Pond water conditioner, increases dissolved oxygen, eliminates suspended solids and reduces ammonia, develops strength and reduces mortality in fish	CAA/JUN 18/FA/01814
Bioyeast	Natural yeast *Saccharomyces cerevisiae*	Feed application: 1-2 kg/ton feed; Pond application:0.808 kg/ha	Releases nutrients for microflora, increases the number of digestive bacteria, improves FCR.	CAA/APR19/FA/03447
LIV-52 (Protek)	Extracts of sarapunka, bhumyamalaki, arjuna, yavatikta, kakamachi, nimba, punarnava, bhringaraja	Fish/shrimp: 20 ml/kg feed (up to 10 weeks age) 50 ml/kg feed (above 11weeks age)	Growth promoter, production enhancer	CAA/MAR 18/FA/01443
Ecomarine	*Bacillius subtilis, Bacillius pumilis, Bacillus amylolichenifaciens, Bacillius megaterium*	40 tablets/ha for 15 days	Absorbs toxic gases, maintains healthy pond bottom, controls harmful blue-green algae	CAA/O16/PRO/00825
Gut plus	Selective beneficial bacteria, vitamins, minerals and enzymes	5-10 g/kg feed for 4 days a week	Activates gut function and provides healthy gut; prevents the infections in gut track; increases appetite and makes low FCR.	CAA/JUL 18A/PRO/02067
Pond max	Blend of naturally occurring bacteria	3.75 g/2500 L of pond water every week for 1 month	Helps to remove pond sludge and excess nutrient levels in the pond, helps to establish a population of good bacteria	CAA/JUL 18/PRO/01953

Multivitamin mix, super mineral, calcium nutria, grow more, vitamin boost, Dmax and probiotica are registered under the Coastal Aquaculture Authority (CAA), India; NA: Not available; FCR: Food Conversion Ratio.

Table 4: Antibiotics and antimicrobial compounds used in freshwater aquaculture farms of West Bengal

Trade name	Active ingredients	Dose and dosage	Indications	Approval status
Terramycin 200, antibac, terramix	Oxytetracycline dihydrate	60mg OTC/kg fish biomass/ day for 10 days	Controls bacterial hemorrhagic septicemia	USFDA (Freshwater-reared salmonids: 3.75 g OTC/100 lb fish/day for 10 days)
Ciproxin	Ciprofloxacin 500	0.94 g/L of water for 7 days interval	Bactericidal	Prohibited for extra-label use by USFDA
Enrocin	Enrofloxacin 10% oral solution	2.5-5.0 mg/L × 5 h bath; 5-10 mg/kg feed for 10-15 days	Controls skin bacterial infections, red disease, ulcers	Prohibited for extra-label use by USFDA
Zithromax	Azithromycin monohydrate	75 g/L of water/day for 5 days	Antibacterial agents	Not approved for fish
Zerobac	Enrofloxacin	10mg/kg body weight for 2days	Controls bacterial infections	Prohibited for extra-label use by USFDA
Erythromycin	Erythromycin A	100 mg/kg feed for 10days	Reduces bacterial growth	Unapproved by USFDA
Bactrim DS	Trimethoprim/sulfamethoxazole	75 mg/kg feed for 8-12 days	Broad spectrum activity	Not approved for fish
Polycure B	Betamethasone, chloramphenicol and polymixin B sulphate	50 mg/kg body weight for 10 days	Reduces inflammation, inhibits bacterial growth	Prohibited for extra-label use by USFDA
Fish pen forte	Penicillin-G potassium	37.8 g/L of water/ for 24 hrs interval	Controls Gram-positive bacteria such as *Streptococcus* spp.	Unapproved by USFDA
Citromax	A mixture of natural organic compounds and lactic ferments with traces of ascorbic acid, palmitic acid,	1g/kg feed/day for 5 days consecutively	Prevents from bacterial and fungal diseases, improves daily weight gain for	CAA/F16/FA/00047 (5g/kg feed/day for 5 days at regular intervals

	mannose, glucose, glycerides, fatty acids, amino acids, citric bioflavonoids, tocopherols and carriers		maximum yield with better survival	on preventive basis)
Amoxstron	Amoxicillin trihydrate	40 mg/kg body weight	Inhibits the activity of transpeptidase enzymes.	Unapproved by USFDA
CIFAX	Chemical formulation	1L//ha; 1st dose: 7 days before stocking of fry in the pond. 2nd, 3rd and 4th doses: once in every quarter from the time of 1st application	Prevents EUS. Antibacterial and antifungal	CIFA institute; cifa.nic.in/
Biosol	Beneficial bacteria and enzymes	10 L or 5 kg/10000m^2 for 3 weeks	Removes high bacterial load from the water body, inhibits the growth of pathogenic bacteria like *Vibrio* spp.	CAA/AUG18/PRO/02453
Bomoclean	Blend of compounds with alkyl dimethyl benzyl ammonium chloride solution,80%	300 mL/ha	Stimulates moulting, inhibits ectoparasitic growth, clears the mucous secretions in fish.	NA

Grow star, redox, starmin and fish min are registered under the Coastal Aquaculture Authority (CAA), India; NA: Not available; USFDA: The United States Food and Drug Administration; CIFA: Central Institute of Freshwater Aquaculture; EUS: Epizootic ulcerative syndrome

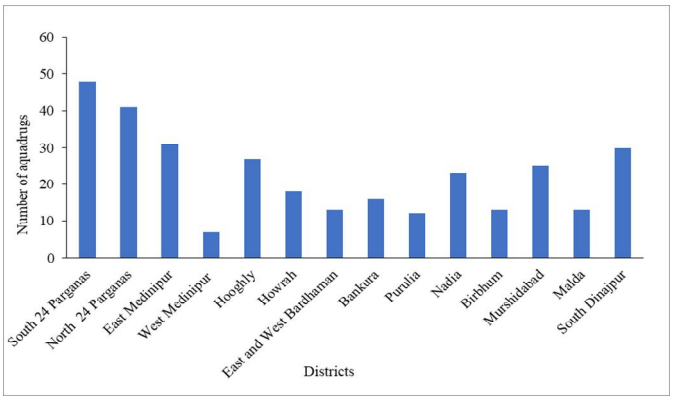

Fig. 2: District-wise usage of aquadrugs in freshwater aquaculture farms of West Bengal

Soil and water treatment compounds

All the respondent farmers from each district reported the use of soil and water treatment compounds during the pond preparation for improving the water and soil conditions (Figs. 3 and 4), with lime (87.28%) being the most commonly used (Fig. 5).

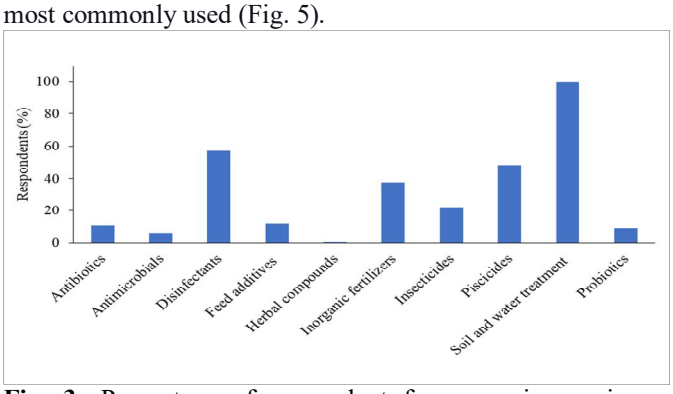

Fig. 3: Percentage of respondent farmers using various aquadrugs in freshwater aquaculture farms of West Bengal

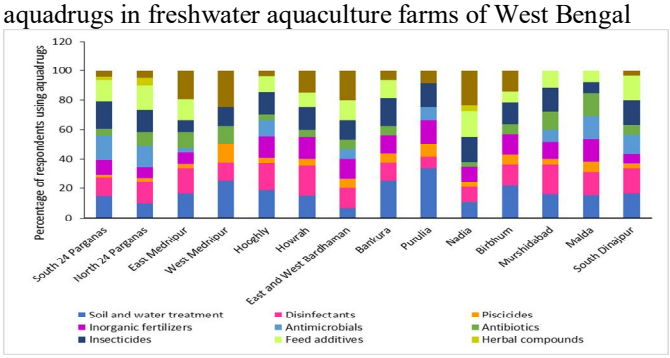

Fig. 4: District-wise usage of major categories of aquadrugs in freshwater aquaculture farms of West Bengal

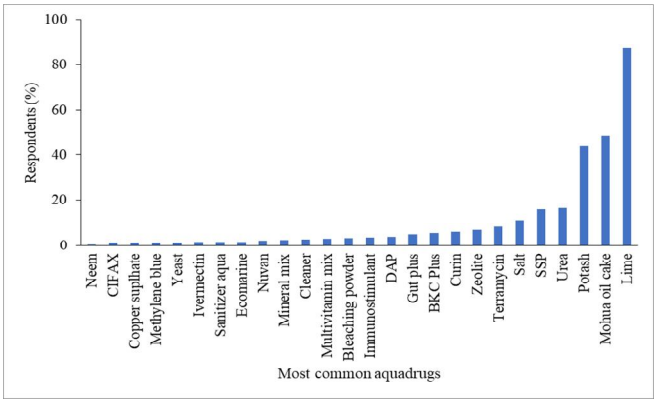

Fig. 5: The most common aquadrugs and chemicals used in freshwater aquaculture farms of West Bengal

These observations are similar to those of earlier studies from other regions (Sharker *et al*., 2014; Mishra *et al*., 2017; Kawsar *et al*., 2019). The application of different lime materials like dolomite, hydrated lime and quick lime was noted in our survey for the improvement of pond productivity. The application of lime may help improve pond productivity, neutralize the acidity of the soil and water, increase total alkalinity and hardness, by providing a source of calcium and magnesium into grow-out ponds (Boyd, 2017; Fitrani *et al*., 2020). Fitrani *et al*. (2020) found hydrated lime tend to be more reliable in terms of effectiveness to minimize the problem in acid sulfate area. Other chemicals like zeolite (6.80%), cure-in (5.92%), Toximar (0.73%), and Aquafresh (0.29%) were also used to increase the alkalinity and remove toxic ammonia from the system (Fig. 5). The use of zeolite and Toximar has been reported commonly for ammonia removal (Ghasemi *et al*., 2018). Likewise, the application of lime, zeolite, insecticides and different fertilizers were common during the pond preparation and water quality management in Noakhali district, Bangladesh (Chowdhury *et al*., 2012). The fish in the pond are susceptible to diseases due to dissolved oxygen (DO) induced stress. To provide oxygen into the stress condition, most of the farmers reported the use of Oxymore plus to maintain the DO levels (Mishra *et al*., 2017).

Disinfectants

The disinfectants were also the most common aquadrugs after the soil and water treatment compounds. Usage of 14 different water disinfectants was documented by 57.40% of the respondents (Figs. 3 and 4). The respondents of Howrah and Murshidabad (20% each), Hooghly (18.52%), East Midnapur and South Dinajpur (16.67%) were the major users of disinfectants to prevent bacterial proliferation. The least users were from Purulia (8.33%) district (Fig. 4). The majority of the disinfectants were used during or before stocking the seeds. Overall, the most commonly used disinfectant in each district was potassium permanganate ($KMnO_4$, 43.79%) followed by benzalkonium chloride (5.33%) and calcium hypochlorite (3.11%) (Fig. 5), with intervals between applications ranged from seven days to months. The disinfectants used by the respondent farmers are summarized in Table 1 along with active ingredients. Rahman *et al*. (2017) reported the use of $KMnO_4$ by 22% of the freshwater fish farmers in Bangladesh. The $KMnO_4$ has been used to treat monogeneans and columnaris disease at 5 mg/L (Tonguthai, 2000), juvenile and adult *Argulus coregoni* for over 30 min with 100% mortality at 10 mg/L (Hakalahti-Siren *et al*., 2008) and helminth parasites at 2.9 mg/L (Zhou *et al*., 2017). The use of other disinfectants or sanitisers such as Sokrena-WS, bleaching powder, Aquakleen, BKC plus, etc were also reported in Indian aquaculture (Mishra *et al*., 2017).

Pesticides and insecticides

A total of 12 pesticide and insecticide compounds were used by the respondents to treat parasites, fungal infections in cultured species, and to kill unwanted organisms in the culture ponds. About 22.04% of the respondents reported the usage of insecticides and pesticides in their farms (Fig. 3). The highest frequency of application was found in South 24 Parganas and Bankura (18.75% each) followed by Nadia districts (17.24%) (Fig. 4). The insecticides and pesticides used by the respondents are presented in Fig. 5. Parasitic diseases like myxoboliosis, argulosis and lernaeosis were treated initially with a salt bath followed by medicated feed containing ivermectin, an anti-parasitic chemical (6.42 and 0.21 g/kg feed, respectively). The salt application was given the preference in most of the farms (11.09%). In extreme conditions, in-feed applications with ivermectin (1.33%) were practised, but not as a combined treatment. Ivermectin is an in-feed antiparasitic veterinary drug that has been approved in the UK. It was successful in reducing the number of chalimus stages of the sea louse, *Lepeophtheirus salmonis* (Horsberg, 2012; Anandaraja *et al*., 2020). Emamectin benzoate is the only chemotherapeutic that has been recommended as an antiparasitic agent under emergency drug release in Canada and investigational new animal drugs (INADs) in the US for the control of ectoparasites such as sea lice infestations in fish (Burridge *et al*., 2010). Most of the surveyed farms of each district used the insecticide dichlorvos (2.07%) directly into the pond to kill the pests entering the ponds with the in-flow water. Ivermectin has a wider efficacy than the organophosphate compounds dichlorvos and azamethiphos and hydrogen peroxide (Davies and Rodge, 2000). Dichlorvos is reported to be the toxic organophosphate to the ecological and economical fish species by several authors (Patar *et al*., 2015; Rao *et al*., 2017). Formalin was also used by many farmers (0.15%). Formalin has been approved by the USFDA for use in aquaculture to treat external trematodes at 250 mg/L for 1 hour (USFWS, 2015) and the treatment dose is strongly dependent on the fish age and water quality during the intensive aquaculture (Leal *et al*., 2016). Overuse of formalin may damage the fish gills, alter the mucosal cells and interfere with the nitrification process (Leal *et al*., 2016). Copper sulphate was also used by 1.04% of the respondents during the culture period to remove the unwanted organisms including the algae. Methylene blue and malachite green were used by 1.18% of the farmers to treat opportunistic fungal pathogen, *Saprolegnia* or oomycete infection in fish. Kawsar *et al*. (2019) reported on the use of methylene blue and malachite green for treating various diseases of fish in Bangladesh.

Antibiotics

Antibiotics may be grouped into natural and synthetic compounds that work by either killing or inhibiting the pathogens (Watts *et al*., 2017; Limbu *et al*., 2020). In West Bengal, about 10.80% of the fish farmers admitted the use of antibiotics like oxytetracycline (OTC), ciprofloxacin, azithromycin monohydrate, trimethoprim and sulfamethoxazole, enrofloxacin, erythromycin, chloramphenicol and amoxicillin trihydrate during the culture to treat a few bacterial diseases (Fig. 3; Table 3). The OTC (8.28%) was the major antibiotic used in each district (Fig. 5), which was administered as a medicated feed to treat several bacterial diseases such as haemorrhagic septicemia, ulcer/red disease, columnaris, gill/tail/fin rot and gill disease. Terramycin-500, a brand name with an active ingredient OTC, is one of the USFDA approved chemotherapeutics for treating specific bacterial diseases in temperate and warm water finfish (USFWS, 2015). The positive effect of OTC oral therapy against *Aeromonas hydrophila* (Julinta *et al*., 2017) and *A. caviae* (Roy *et al*., 2018) infection was demonstrated in *Oreochromis niloticus*. The dose and dosage of antibiotics used by the respondents during the culture period are tabulated in Table 4. The respondents using the antibiotics was the maximum in Malda (15.38%) followed by West Midnapur (12.50%), Murshidabad (12.00%), East Midnapur (11.11%), North 24 Parganas (9.76%), Birbhum (7.14 %), East and West Bardhaman and South Dinajpur (6.67% each), Bankura (6.25%), Howrah (5.00%), South 24 Parganas (4.17%), Hooghly (3.70%) and Nadia (3.45%) districts (Fig. 4). None of the Purulia farmers admitted the use of antibiotics during the culture period. In contrast, Abraham *et al*. (2018) reported the use of terramycin and pentamycin by 27% of the African catfish farmers in North and South 24 Parganas districts of West Bengal. Rico *et al*. (2013) reported the use of antibiotics in 100% of the *Pangasius hypophthalmus* farms followed by *O. niloticus* farms in China and Thailand; while antibiotics usage was the lowest in Bangladesh. Recently, Lulijwa *et al*. (2020) documented the use of 67 antibiotic compounds in 15 major aquaculture producing countries between 2008 and 2018. A global survey by Tusevljak *et al*. (2013) revealed that antibiotics are widely applied in aquaculture in Asia, Europe, the USA, Canada, and other countries, which seriously impacted food safety in terms of the residues of antibiotics (Chen *et al*., 2015, 2018). In Bangladesh, Kawsar *et al*. (2019) reported

14 antibiotic compounds to cure the different fish bacterial diseases. According to them, OTC and erythromycin were used by 10 and 52% of the farmers, respectively. It has been reported that the non-metabolized antibiotics that are consumed through feed or as medicine directly enter into the environment through faecal matters, leading to the development of antibiotic resistance in aquatic bacteria (Watts *et al*., 2017). Antimicrobial resistance is a major public health crisis by overuse or misuse of antimicrobials in aquaculture that can compromise the treatment of bacterial infection (Ferri *et al*., 2017; Love *et al*., 2020).

Antimicrobials

Most antimicrobials are used for the prophylactic measure in aquaculture. Twenty-three different antimicrobial compounds were used by 5.92% of the respondents (Fig. 3) for disease management. The respondents from the South 24 Parganas district were the highest users of antimicrobials (16.67%) followed by Malda (15.38%), North 24 Parganas (14.63%), South Dinajpur (13.33%) and Hooghly (11.11%) districts. The West Midnapur, Howrah and Birbhum districts recorded the least usage (Fig. 4). The CIFAX was the most commonly used antimicrobial compound, i.e., 1.04% of the respondents to prevent ulcerative diseases (Fig. 5). Other antimicrobial products like Stop star, Blitox 50, Redol, Geodon, Hexidol, Dexdomitor, Lexyncet, etc have been used by the respondent farmers. The results corroborate the observations of Mishra *et al*. (2017).

Piscicide

Mahua oil cake (MOC), a derivative from the plant *Madhuca longifolia*, is widely used in aquaculture practices as a piscicide. The MOC contain about 7.8-8% saponin, an active ingredient, that acts as a fish toxicant and kills the predatory fish. About 48.52% of the respondents reported the use of MOC (Figs. 3 and 5). The MOC also act as organic manure in fish ponds after its toxic effects that help to enrich with balanced nutrients (Vinothkumar *et al*., 2018).

Fertilizers

Fertilization usually results in a 2-5 folds increase in aquaculture production and the combination of two types of fertilizers results in much greater production (Boyd, 2018). The respondent fish farmers of West Bengal reported the usage of organic and inorganic fertilizers in their farms, however, the dose varied. The inorganic fertilizer application was common among the respondents (37.72%) besides the use of soil and water treatment compounds, disinfectants and piscicides to raise fish production. The number of respondents using the inorganic fertilizers was almost the same in most of the districts, except for South Dinajpur (6.67%) (Fig. 4). The most common inorganic fertilizers were urea, phosphate and single superphosphate (Fig. 5). The use of organic manure (cattle manure) was reported by only 16.86% of the respondents. Average daily inputs of organic manure have shown a greater influence on the net fish production in ponds to about 150 kg/ha/day (Knud-Hansen *et al*., 1991; Boyd, 2018).

Feed additives and herbal compounds

The feed is one of the important components in aquaculture that constitute 50-60% of the total operational cost (Mishra *et al*., 2017). Various essential additives are incorporated at required levels to the basic formulation to achieve balanced nutrients. During the survey, it was observed that farm-made feeds such as rice bran (40.83%), mustard oil cake (11.39%) and groundnut oil cake (0.15%) were used by many farmers. About 11.98% of the respondents reported the use of feed additives and a few for plant products (0.59%) (Fig. 3). The use of essential additives such as vitamins and micro and macro minerals was noted. Multivitamins (2.96%) and mineral mix (2.37%) were the commonly used feed additives by the fish farmers of West Bengal (Fig. 5). Many growth-promoting non-essential additives like enzymes, Aquazyme, *Saccharomyces* spp. etc were also used by the respondents. The use of various enzymes like Polzyme, Biozyme and Acmezyme and other products (Megavit Aqua, Aqua Boost, Aqua Savor, Vitamin premix, Fibosoel, Grow fast, etc) by the Bangladesh fish farmers have been reported (Sharker *et al*., 2014; Rahman *et al*., 2017; Kawsar *et al*., 2019). The major users of feed additives were from Nadia (17.24%) followed by North 24 Parganas (17.07%), South Dinajpur (16.67%) and South 24 Parganas (14.58%) districts. The least usage of feed additives was noted in Malda (7.69%) and Birbhum (7.14%) (Fig. 4). Mishra *et al*. (2017) also reported the use of a wide range of feed supplements like Liv 52, Protec, Agrimin, Superfood, essential amino acids, etc by the farmers and hatchery operators in India. About 3.55% of the respondents used immunostimulants as dietary additives to improve the growth and survival rates by enhancing the non-specific defence mechanisms and increase resistance to specific pathogens (Fig. 5). The common herbal compounds used were turmeric (*Curcuma longa*) and neem (*Azadirachta indica*).

Probiotics

The main interest of probiotics is as growth promoters and to improve the health of fish and as a stress tolerance (Hoseinifar *et al*., 2018). In this survey, about 8.88% of respondents reported the application of in-feed probiotics and soil and water probiotic products in their farms (Fig. 3). The respondents of West Midnapur (25.00%), Nadia (24.14%), East and West Bardhaman (20.00%) and East Midnapur (19.44%) were the common users of probiotics. The least users were from the South Dinajpur (3.33%), Hooghly (3.70%) and South 24 Parganas (4.17%) districts (Fig. 4). Soil and water probiotics used in the surveyed farms were Aqua Tab, Super Plankt, Bottom clear, Metaprob, Promax, Zeepromin, Ecomarine, Probio Aqua, Protab and Pond fresh. The in-feed probiotics like Y-max, Lacto-plus, Gut-plus, Growth-plus, Probiotica, Lactomin were also used by the farmers. Gut-plus and Ecomarine were the commonly used probiotic products by 4.88% and 1.33% of the farmers, respectively (Fig. 5). These products contained microbiota like *Lactobacillus* spp., *Bacillus* spp., *Nitrosomonas* spp., *Nitrobacter* spp., *Rhodobacter* spp., photosynthetic bacteria and yeast for the nutritional and enzymatic contribution to digestion and health conditions of the cultured species and water quality improvement. Likewise, Mishra *et al*. (2017) also noted the usage of many

probiotics mostly in shrimp culture practices and carp farms.

Each product that has been reported by the fish farmers of West Bengal during the study period has been verified from authentic sources such as CAA and USFDA, whether the aquadurgs and chemicals are approved or unapproved or registered as antibiotic-free products and the details are presented in Tables 1-4. There are national/regional maximum residue limits (MRLs) for several antimicrobials and veterinary drugs used in aquaculture (Karunasagar, 2012). In our study, aquadrugs and chemicals banned under national and international regulations were also found to be used by the interviewed farmers. Chloramphenicol and nitrofurans are banned for use in food-producing animals in most Asian countries (Karunasagar, 2012; Rico *et al*., 2012). Malachite green, a pesticide/fungicide, has been banned in many countries (Rico *et al*., 2012). In 4 out of 676 surveyed farms of West Bengal (South 24 Parganas (1), North 24 Parganas (1) and Howrah (2) districts, the use of malachite green was noted so also several other pesticides.

Fish production and aquadrugs usage per ton fish production

The respondent farmers (n=676) reported fish production in the range of 1,000 kg/ha/year in South 24 Parganas to 18,043 kg/ha/year in East and West Bardhaman districts, with an average fish production of 5,827±3364.83 kg/ha/year (Table 5). The estimated quantity of each aquadrug, chemical or probiotic product/ton fish production in West Bengal is provided in Table 6. The input in terms of quantity was dominated by MOC (233.19± 189.87 kg/ton fish production). The MOC was used for the removal of unwanted fish entering the ponds with the in-flow water, to control the rise in mortality and to avoid the usage of harmful synthetic products, which leave residues in the aquatic environment (Vinothkumar *et al*., 2018). The inputs of soil water treatment products were the next dominant and these included lime (48.70±46.62 kg/ton fish production), Toximer (40.67±20.63 kg/ton fish production) and zeolite (20.76±18.25 kg/ton fish production). The use of inorganic fertilizers was 27.04±24.19 kg/ton fish production in the surveyed fish farms. Among the disinfectants, the use of bleaching powder (2.90±2.77 kg/ton fish production), benzalkonium chloride (2.70±2.62 L/ton production), $KMnO_4$ (1.07±1.01 mg/ton fish production) and iodine (0.35±0.33 L/ton fish production) were common. The products related to parasiticides, pesticides and insecticides were dominated by water broadcasting products such as nuvan and formalin (17.62±9.33 mL/ton fish production) and in-feed incorporation compound, ie., ivermectin (1.33±1.29 g/ton fish production). The application was mostly to control *Myxobolus* spp., *Argulus* spp. and *Lernaea* spp. infestation. Except for formalin, the products are not recommended or permitted for use in aquaculture (USFWS, 2015).

Usage of antimicrobials in food animal production collected by surveillance, monitoring, and reporting is an important component of national antimicrobial resistance policies, as antimicrobial use is proportional to antimicrobial resistance (WHO, 2017; Love *et al*., 2020). In West Bengal, the application of antibiotics (0.35±0.23 L/ton fish production and 4.59±2.13 mg/ton fish production) was the least. The medicated feeds were the highest contributors (1.38±1.34 g/ton fish production) followed by dip treatment antibiotic products (4.59±2.13 mg/ton fish production). On the other hand, antibiotics usage to the tune of 580 g, 175 g and 93 g/ton harvested fish respectively in Chile, Canada and Vietnam, particularly in salmon and *Pangasisus* farms (Rico *et al*., 2013) was documented. According to Lulijwa *et al*. (2020), 73% of the farms in 11 major fish producing countries applied OTC, sulphamethoxine, erythromycin, amoxicillin and enrofloxacin. It has been reported that approximately 80% of the antimicrobials used in aquaculture enter the environment through uneaten medicated pellets and faeces (Cabello *et al*., 2013; Love *et al*., 2020). In our survey, the application of antibiotics in feed was observed to be within the recommended dosage, i.e., 2-4 g OTC/kg feed (Julinta *et al*., 2017).

Among the feed supplements, the major contributors were the minerals as Aquamin (3.43±1.21 kg/ton fish production followed by multivitamins (3.82±3.31 g/ton fish production) and growth promoter enzymes (3.12±1.49 mL/ton fish production). The freshwater aquaculture farms generate high concentrations of unutilized nitrogen that may contribute to the disease occurrence (Boyd, 2017). The soil and water probiotics were the maximum contributors among the probiotics to improve the water quality by removing the high concentrations of nitrogen production and toxic ammonia from the aquaculture system and the usage was 3.89±3.63 kg/ton fish production. The usage of feed probiotics to improve the health of cultivated species was 2.98±2.75 g/ton of harvested fish production. The medicinal plants play a great role as a growth promoter, immunostimulants, appetite stimulator, antimicrobial and anti-stress agents and also acts as an aphrodisiac (Citarasu, 2010). The average usage of the herbal compounds was 1.79±1.68 g/ton harvested fish production. The dietary herbal products have shown improved growth in fish (Ashraf and Goda, 2008).

Table 5: Aquacultured fish production in the surveyed districts of West Bengal

District (Number of respondents = 676)	Fish production in kg/ha/year		
	Minimum	Maximum	Mean±SD
South 24 Parganas (n=90)	1000	14911	6785.80±3861.23
North 24 Parganas (n=61)	1812	16086	6892.26±3382.64
East Midnapur (n=52)	1995	14864	4251.79±2889.17
West Midnapur (n=19)	1932	7497	4088.50±2551.88
Purulia (n=27)	2211	7194	4158.11±1796.25
Bankura (n=31)	3155	17690	7501.90±3821.17
Birbhum (n=7)	1846	10620	5695.60±3537.74
E&W Bardhaman (n=91)	1914	18043	6561.10±4188.30
Howrah (n=50)	1573	17891	7350.42±4044.09
Hooghly (n=90)	1901	15091	5549.46±3477.98
Nadia (n=82)	1821	17635	7365.44±4104.28
Murshidabad (n=29)	1726	16820	6008.84±3625.37
Malda (n=27)	1910	8522	4528.12±2022.93
South Dinajpur (n=20)	1929	5775	4837.10±3804.60

*Average fish production in the surveyed fish farms of West Bengal (n-676) was 5,827±3364.83 kg/ha/year. SD: Standard deviation

Table 6: Aquadrugs usage per ton freshwater fish production in West Bengal

Aquadrugs	Usage/ton fish production		
	Minimum	Maximum	Mean±SD
Lime (kg/ha)	5.15	387.10	48.70±46.62
Disinfectant (L/ha)	0.19	12.87	2.70±2.62
Disinfectant (kg/ha)	0.08	12.87	2.90±2.77
Pesticides and insecticides (g/kg feed)	0.21	6.42	1.33±1.29
Pesticides and insecticides (mg/L)	2.15	128.71	41.32±37.70
Pesticides and insecticides (mL/ha)	4.72	36.30	17.62±9.33
Mohua oil cake (kg/ha)	12.83	1795.67	233.19±189.8
Probiotic (kg/ha)	0.42	12.90	3.89±3.63
Probiotic (g/kg feed)	0.08	7.72	2.98±2.75
Immunostimulant (g/kg feed)	32.18	170.76	112.13±46.70
Herbal compound (g/kg feed)	0.11	4.29	1.79±1.68
Inorganic fertilizer (kg/ha)	2.57	128.71	27.04±24.19
Antibiotics (g/kg feed)	0.13	5.15	1.38±1.34
Antibiotics (mg/L)	2.57	6.44	4.59±2.13
Antibiotics (L/ha)	0.09	0.64	0.35±0.23
Antimicrobial (CIFAX) (L/ha)	1.46	1.75	1.60±0.21
Antifungal (mg/L)	0.11	3.09	1.28±1.08
Algaecide (mg/L)	1.95	4.29	3.12±1.65
Minerals (g/kg feed)	0.02	4.29	1.97±1.52
Minerals (kg/ha) (Aquamin)	2.57	4.29	3.43±1.21
Feed supplements (g/kg feed)	0.16	12.87	3.82±3.31
Feed supplements (mL/kg feed)	1.72	2.57	2.15±0.61
Vitamins (g/kg feed)	0.09	6.01	1.71±1.24
Enzymes (mL/kg feed)	1.72	5.41	3.12±1.49
Potassium permanganate (mg/L)	0.09	5.32	1.07±1.01
Iodine (L/ha)	0.03	1.29	0.35±0.33
Toximer (kg/ha)	24.88	64.01	40.67±20.63
Zeolite (kg/ha)	5.16	60.07	20.76±18.25

CONCLUSION

The present study documented the current status of the usage of aquadrugs, chemicals and probiotic products in aquaculture in West Bengal. The quantity of aquadrugs usage was, however, far less than those recorded in many other major fish producing countries. Though the aquadrugs and chemicals are important components for successful aquaculture practices, the majority of the farmers lacked knowledge on the chemicals, doses and methods of application of these chemicals. With the further development of the aquaculture industry, particularly in the intensified system, the applications of drugs and chemicals would be increased. The farmers must have access to a range of authorized medicines and technical inputs to safeguard fish health. Also, they must be educated on the use of authorized medicines, environmental impacts, national or international standards on drug residues and consumer safety. The researchers, policymakers and extension workers should work together to address these issues in aquaculture with the view to reduce the negative impacts and for sustainable aquaculture.

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AUTHORS' CONTRIBUTIONS

JS, AR, GD and TSN participated in the primary data collection; TJA and PKP conceptualized, generated funding and lead the data collection; JS wrote the draft manuscript. TJA developed the manuscript structure, edited and finalized the manuscript

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