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Antibiotic Residues in Seafood – A Hazard

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Wild shrimps, being mainly marine, have no chance of having antibiotic residues. But farmed shrimp used to have antibiotic residues as a consequence of aquaculture practices. Antibiotics used in aquaculture, whether as therapeutic, prophylactic or growth promoter will accumulate in shrimp tissues and exoskeleton. International trade regulations do not permit antibiotic residues in processed fishery products. Instances of rejection/detention of frozen shrimp/scampi in the port of entry of importing countries, due to detection of antibiotic residues are very frequent. The paper reviews the present status of antibiotic residues in shrimp vis-à-vis the trade regulations and discusses the problems faced by seafood processors.

Key words : Antibiotic residues, aquaculture drugs, environment, health hazards

Antibiotic residues in processed fish and fishery products have become a serious health hazard to the consumer and an economic hazard to the exporting nations. Now this is the major quality problem being faced by the Indian fish farmers and exporters. The processed shrimp exported from India includes wild shrimp from both the sea and brackish water and farmed tiger prawn (*Penaeus monodon*) and scampi (*Macrobrachium rosenbergii*). Wild shrimps are mainly marine and only a very small share comes from open brackish water areas. Marine ecosystem is free from antibiotics and hence there is no chance for the presence of antibiotic residues in marine shrimps. But farmed shrimps can have antibiotic residues as a consequence of aquaculture practices.

Even though the use of antibiotics in aquaculture practice is unscientific, unwanted and harmful, they are being used for (i) therapeutic (ii) prophylactic and/or (iii) growth promoting purposes. Also, some manufactures are incorporating certain antibiotics in shrimp feed as a feed preservative. The devastating shrimp diseases like white spot syndrome and yellow head disease are caused by viruses. Antibiotics have no therapeutic value against viruses at all. Still many of our shrimp farmers are dumping antibiotic formulations in their farms against viral diseases. Further, even bacterial diseases cannot

be treated with antibiotics, since in the aquaculture environment, effectiveness of antibiotic therapy is not at all proved

Antibiotics should not be used in aquaculture because of the following reasons:

(i) Effect on environmental microflora

The aquaculture environment is a very dynamic system. The chemical/biochemical/gaseous equilibrium in the farm, i.e., the water quality, is maintained by the mineralisation process caused by microflora, mainly bacteria. In the shrimp farms, on an average, 60 $mg.m^{-2}$ of waste is produced for every kilogram of shrimp produced. These organic wastes are mineralized and farm environment kept clean and healthy by the farm/soil microorganisms. When we use antibiotics/antibacterial chemicals in the farm either as therapeutical, prophylactic or growth promoters, they destroy the environmental microflora. The scavenging action by microflora is stopped. So waste accumulates in the farm, resulting in a favourable environment for disease outbreaks.

(ii) Development of drug resistant bacteria, including pathogens.

Use of antibiotics results in the emergence of drug resistant bacteria, some of which are human pathogens.

(iii) Retention of drug residue in farmed shrimps

The antibiotics used in aquaculture will accumulate in the shrimp tissue and exoskeleton. The drug so accumulated has to be eliminated from the tissue by a biological process called detoxification. In the case of aquatic animals, the biochemical process of drug elimination is very slow compared with land animals. The drug withdrawal period before harvesting, usually 2-3 weeks will not eliminate the accumulated residues from the tissues, particularly from exoskeleton. So, the accumulated antibiotic residues will remain in tissues and exoskeleton, which will be detected in the testing laboratory. The European Union (EU, 1990), USFDA (USFDA, 2001) and Japan have notified that residues of the antibiotics listed in their notification should not be present in the imported shrimp.

(iv) Health hazard to the consumers

Antibiotics are used as therapeutics in humans to fight microbial diseases. But, certain antibiotics have been shown to cause serious health

hazards. So, antibiotic residues in food can be dangerous to the consumer. For example, chloramphenicol is known to cause haemotoxic side effects, particularly the chloramphenicol induced aplastic anaemia, which is often fatal. Nitrofurans and their metabolites are genotoxic and carcinogenic. So far, no dose-effect relation could be established in man for these two groups of antibiotics. So, they are declared as zero tolerant antibiotics.

Table 1. Permitted maximum residual level (MRL) of antibiotics in seafood as notified by EU

Antibiotic	MRL permitted in food/fish products*	
Sarafloxacin	0.03 ppm	Salmonid fishes
Nafcillin	0.3 ppm	Meat
Nafcillin	0.03 ppm	Milk

* No other antibiotics are permitted in food products

European Union standards (EU, 1990) stipulates that anti-infection agents, antibiotics and quinolones should not be present in fish/meat products imported to the EU countries except those mentioned in Table 1. Permitted maximum residue levels of notified antibiotics as per USFDA (2001) are given in Table 2 and those by the Government of India (GOI, 2001), in Table 3. Japan permits no antibiotic residue except tetracyclines, to a maximum of 0.1 ppm in seafood.

Table 2. Permitted MRL of antibiotics in seafood as per USFDA (2001)

Antibiotic		MRL permitted in seafood
Oxytetracycline	2 ppm	In salmonids, cat fish and lobster only
Sulfamerazine	Nil	All fishes
Sulphadimethoxine/Ormetoprim combination	0.1 ppm	In salmonids and cat fish only
Other antibiotics	-	Not permitted in fish/fishery products

Present scenario in India

Antibiotics and antibacterial substances are indiscriminately being used in India in shrimp farms in Andhra Pradesh, Tamil Nadu, Kerala and Karnataka. Maximum use takes place in Andhra Pradesh. A recent study in Central Institute of Fisheries Technology, Cochin showed that out of the 2086 samples of farmed shrimp (tiger prawn, white prawn and freshwater

scampi) tetracyclines were detected in 134 samples and chloramphenicol in 28 samples. Recently, chloramphenicol and nitrofurans were detected in Indian farmed shrimp exported to Spain, Netherlands and UK in ppb levels. All the consignments were confiscated and destroyed. Indian Exporters are reported to have lost nearly Rs.100 crores. The Government of India, Ministry of Agriculture (GOI, 2002) have banned the use of 24 antibiotics and pharmacologically active substances in aquaculture (Table 4).

Table 3. Maximum Residual Level (MRL) of permitted antibiotics by Government of India in fishery products (GOI, 2001)

Antibiotics	MRL, ppm
Chloramphenicol	Nil
Furazolidone	Nil
Neomycin	Nil
Tetracycline	0.1
Oxytetracycline	0.1
Oxolinic acid	0.3
Trimethoprim	0.05
Nalidixic acid	Nil
Sulphamethazole	Nil

For export to EU, USA and Japan, the MRL fixed by individual countries are to be complied with.

EU perception of the residues problem in seafood

The EU has issued Council Regulation 2377/90, updated up to 1-12-2000 enlisting nearly 300 pharmacologically active substances/antibacterials/ antibiotics/sulphonamides, which are either declared zero tolerant or for which maximum residue limits (MRL) have been fixed. But, the MRLs are less than 50 ppb in most of the cases. The following antibiotics are banned in seafood (all foods) vide EU legislation 96/23/EC & EEC no2377/90 (EEC, 2002).

i. Chloramphenicol
ii. Nitrofurans:

Nitrofurazone
Nitrofurantoine
Furazolidone
Furaltadone and their metabolites

Further, maximum residue limits (MRL) have been fixed for the antibiotics as given in Table 5.

Table 4. Antibiotics and other pharmacologically active substances banned in aquaculture practice in India*

Sl. No.	Antibiotics and other pharmacologically active substances
1	Chloramphenicol
2	Nitrofurans including: Furaltadone, Furazolidone, Furylfuramide, Nifuratel, Nifuroxime, Nifurprazine, Nitrofurantoin, Nitrofurazone
3	Neomycin
4	Tetracycline
5	Oxytetracycline
6	Trimethoprin
7	Oxolinic acid
8	Nalidixic acid
9	Sulphamethoxazole
10	Aristolochia spp and preparations thereof
11	Chloroform
12	Chlorpromazine
13	Colchicine
14	Dapsone
15	Dimetridazole
16	Metronidazole
17	Ronidazole
18	Ipronidazole
19	Other nitroimidazoles
20	Clenbuterol
21	Diethylstilbestrol (DES)
22	Sulfonamide drugs (except approved sulfadimethoxine, Sulfabromomethazine and Sulfaethoxypyridazine)
23	Fluroquinolones
24	Glycopeptides

* The list is proposed to be included in the amendment to the Notification SO 792 (E) dated August 17, 2001 of the Ministry of Commerce and Industries

Table 5. Maximum residue limits (MRL) for antibacterials and antibiotics in EU

Antibacterials/Antibiotics	MRL, ppb
Trimethoprim	50
Beta lactams (Penicillins)	4 to 300
Cephalosporins	20 to 1000
Quinolins	50 to 1500
Macrolids (Tylosin group)	50 to 200
Tetracyclines	100 to 600
Aminoglycosides (Streptomycins)	100 to 1000

1 ppb = 0.001 ppm

Difficulties in analytical field

The assay of antibiotics in tissues is posing great difficulties in analytical laboratories. So far, the microbiological assay as per AOAC (2000) methods, using specific sensitive bacterial cultures has been in vogue everywhere. But the microbiological assay has the limitation of detection level. At present, under the best conditions and using the most sensitive strains, the lowest limit of detection is only 100 ppb (i.e., 0.1 ppm) at 95% confidence level. Hence the microbiological assay will not detect zero tolerant antibiotics, if less than 100 ppb.

Instrumental methods like HPLC and GC have also the limitation of detection levels at present being only 100 ppb. Recently HPLC-MS-MS (LC-MS-MS) and GC-MS have come to the scene, which could detect in ppb levels. However, the cost involved in the analytical set up is enormous. Now, ELISA kits are available for the detection of certain antibiotics like chloramphenicol. Some of the kits have claimed to have a minimum detection level of 0.025 ppb of chloramphenicol in seafood.

Difficulties for seafood processors

More than 50% of the processed shrimps including scampi is of aquaculture origin. Testing facilities for detection of very low levels of antibiotic residues in the raw material, are not available in seafood processing establishments. Hence, seafood processors have to depend on the declarations from the farmers/suppliers to the effect that no antibiotics have been used in the farm. When the residues are detected in the importing countries, the seafood processors stand to lose their money and reputation as supplier of safe products.

How we will redress the problem?

Only earnest and concerted action at the following levels will redress the antibiotic residue problem in Indian seafood:

i. Action at Government level to ensure that the banned chemicals/ antibiotics are not available to farmers and are not used in farms. The following Government Departments may be given the responsibility for this:

   Central - Agriculture, Commerce and Environment Ministries
   State - Fisheries and Environment Departments

ii. Action at farmers' level - No aquaculture drug to be used in farm.

iii. Action at buyers' level (seafood processors) - to be vigilant on the presence of drug residues in raw materials

iv. Action at Inspection/Enforcement/Promotion/Research Agencies' level:

Export Inspection Council of India (EIC)

Marine Products Export Development Authority (MPEDA)

Central Institute of Fisheries Technology (CIFT)

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