

PREVENTION OF ANTIBIOTIC RESIDUES IN FARMED SHRIMPS: A STUDY ON DEVELOPMENT OF HACCP FRAMEWORK APPLICABLE AT SHRIMP FARMS IN INDIA

Thesis submitted in partial fulfillment
of the requirements
for the degree of

Ph.D. (INLAND AQUACULTURE)

by

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(IAC-155)**

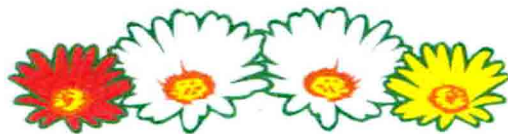


CENTRAL INSTITUTE OF FISHERIES EDUCATION
(Deemed University)

Indian Council of Agricultural Research
Fisheries University Road
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AUGUST 2005

Dedicated
to the everloving
memory of
my most beloved
grandmother
Late Mrs. T. Santhosamma





केन्द्रीय मात्स्यिकी शिक्षा संस्थान

(समतुल्य विश्वविद्यालय) भारतीय कृषि अनुसंधान परिषद

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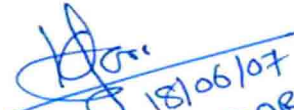


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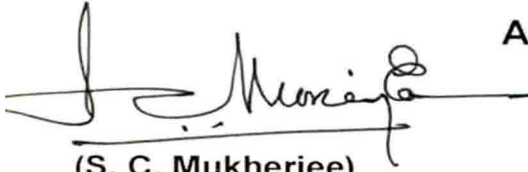
Certified that the thesis entitled "**PREVENTION OF ANTIBIOTIC RESIDUES IN FARMED SHRIMPS: A STUDY ON DEVELOPMENT OF HACCP FRAMEWORK APPLICABLE AT SHRIMP FARMS IN INDIA**" is a record of independent *bona fide* research work carried out by **Mr. Sumanth Kumar Kunda** during the period of study from September 2001 to August 2005 under our supervision and guidance for the degree of **Doctor of Philosophy (Inland Aquaculture)** and that the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or any other similar title.

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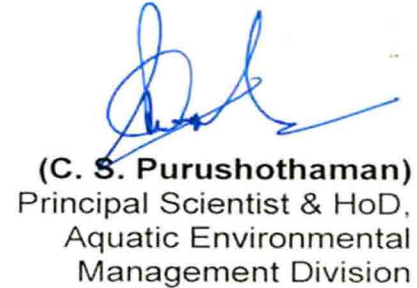

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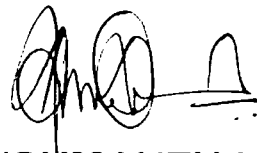

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DECLARATION

I hereby declare that the thesis entitled “**PREVENTION OF ANTIBIOTIC RESIDUES IN FARMED SHRIMPS: A STUDY ON DEVELOPMENT OF HACCP FRAMEWORK APPLICABLE AT SHRIMP FARMS IN INDIA**” is an authentic record of the research work done by me and that no part thereof has been presented for the award of any degree, diploma, associateship, fellowship or any other similar title.

31st August 2005

Mumbai



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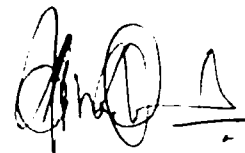
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Mumbai,
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(Sumanth Kumar Kunda)

सारांश

विश्व में जलकृषि खाद्य उत्पादन प्रणाली के रूप में सबसे तेजी से विकसित हुआ है। पिछले दशक में इसका वृद्धि दर 9.6% प्रति वर्ष रहा है तथा वर्तमान में इसका उत्पादन विकासशील देशों में निरंतर बढ़ती जा रही है। भारत श्रिंप के पालन में विश्व में पांचवें स्थान पर है जो अपनी हिरसेदारी लगभग 60.5% का रखती है तथा यह कुल निर्यात मूल्य के 82% के लगभग है। श्रिंप पालन भारतीय जलकृषि उद्योग में एक महत्वपूर्ण अवयव के रूप में उभरकर आया है तथा 75% पालन क्षेत्रों में पीनियस मोनेडोन प्रजाति की खेती (पालन) की जा रही है। विश्व व्यापार संगठन (WTO) की जा रही है। विश्व व्यापार संगठन (WTO) की स्थापना के साथ ही तथा SPS समझौता एवं स्वास्थ्य पहलुओं से संबंधित जागरूकता के साथ भारत में श्रिंप पालन थोड़ा बहुत प्रभावित हुआ है जिसका मुख्य कारण प्रमुख आयातक देश जैसे EU, USA एवं जापान द्वारा कुछ श्रिंप कंसाइनमेंट को अस्वीकार कर देना है। इसके अस्वीकार करने का आधार अप्रतिजैविकी अवशेष की उपस्थिति को प्रतिबंधित करना है। भारत विश्व व्यापार संगठन का एक सदस्य देश है तथा यह विश्व बाजार में भारतीय श्रिंप उत्पाद को बढ़ावा देता है इसलिए इसे अन्तर्राष्ट्रीय गुणवत्ता मानक को बनाए रखने के अलावा कोई दूसरा विकल्प नहीं है। हेज्जार्ड एनालेशिस क्रिटिकल कंट्रोल पॉइंट एक रिस्क मैनेजमेंट प्रोग्राम है जिसे विश्व के अधिकांश देशों में खाद्य सुरक्षा आपदा के रूप में अपना रखा है। इसके साथ ही खाद्य सुरक्षा मुद्दे HACCP सिद्धान्त वैज्ञानिक रूप से आधारित संरचना को प्रदान करता है जिसे रिस्क मैनेजमेंट प्रोग्राम के विकास हेतु उपयोग किया जा सकता है जिसमें बहुत से मुद्दों का उल्लेख किया गया है। इन सब बातों को ध्यान में संचालित किए गए हैं जो मुख्य रूप से भारत के विभिन्न राज्यों के तटीय जिलों से संबद्ध रखते हैं जैसे आन्ध्र प्रदेश, तमिलनाडु, गुजरात, महाराष्ट्र, कर्नाटक एवं केरल है। जहाँ संभावित आपदाओं का विशेषण एवं वर्तमान पालन प्रणाली जो प्रतिजैविकी के उपयोग हेतु उत्तरदायी है का अध्ययन किया जा रहा है। भारतीय श्रिंप पालन में स्थलों का चयन, तालाबों की तैयारी, पानी का भंडारण, श्रिंप बीज, आहार, पानी की गुणवत्ता, प्रतिजैविकी का प्रयोग, फार्म प्रबंध प्रणाली एवं उसका फसलोपरान्त उत्पादन आदि नौ क्रिटिकल कंट्रोल पॉइंट (चिन्हित) पहचाने गये हैं। वर्तमान अध्ययन में, केवल एक किसान को छोड़कर कोई भी प्रतिजैविकी का उपयोग नहीं किया। परिणाम यह दर्शाता है कि पूर्वी तट के 56.9% तथा पश्चिमी तटीय किसानों ने 38.4% WSSV की समस्या का सामना किया है। अधिकांश किसान बीज गुणवत्ता (23.3%) जलवायु (20.8%) एवं पानी की गुणवत्ता (19.1%) जलवायु (20.8%) एवं पानी की गुणवत्ता (19.1%) के कारण अपने फसल की हानि सही है। आन्ध्र प्रदेश (13.3%) एवं कर्नाटक (53.8%) के किसानों की समुदायों ने लूज शेल सिन्ड्रोम की समस्या को झेल है जबकि अधिकांश किसान अच्छे फार्म प्रबंध से संबद्ध हैं जिसके कारण सफलतापूर्वक पालन कर रहे हैं। कुल जबाबकर्ताओं में से लगभग 30% ने यह सुझाव दिया कि इस हेतु आर्थिक सहायता एवं प्रयोगशाला सुविधाएं उपलब्ध कराने पर श्रिंप पालन में अच्छे परिणाम आएंगे। प्रतिजैविकी एवं जल स्रोत के उपयोग के बीच ऋणात्मक सहसंबंध 5% स्तर तक था तथा प्रोबायोटिक, शिक्षा स्तर एवं जल स्रोत के इस्तेमाल के बीच धनात्मक सहसंबंध 1% के स्तर तक सार्थक था। पहचाने गए CCP के लिए क्रिटिकल लिमिट को फिक्स करना तथा प्रयोगात्मक परीक्षण से संबंधित कंट्रोल मापदण्ड का प्रयोग किया गया। यह प्रयोग ऑक्सीट्रेटासाइक्लिन के लिए निकासी अवधि का आकलन करना था। नमूनों को HPLC के इस्तेमाल एवं UV detector के इस्तेमाल से विश्लेषण किया गया। परिणाम यह दर्शाता है कि श्रिंप के शरीर से किसी भी प्रतिजैविकी का पूर्ण निष्कासन के लिए 21 दिनों की अवधि अपेक्षित है। ANOVA परिणाम यह संकेत करता है कि नियंत्रण एवं उपचार के बीच कोई उल्लेखनीय अन्तर नहीं है। व्यवसायिक श्रिंप आहार के दस विभिन्न ब्रांड आन्ध्र प्रदेश से इकट्ठा किया गया एवं उसका सूक्ष्म जैविकी विश्लेषण किया गया इस विश्लेषण में यह देखा गया कि ओलीफार्म की गिनती का रेंज 1×10^2 से 4.2×10^2 cfu/g एवं कुल प्लेट कॉन्ट्रेंज 1×10^4 से 3.2×10^3 cfu/g देखा गया। भौतिक बाधा यह प्रकट करता है कि श्रिंप आहार के अल्पसंरचना तथा अधिकांश आहार नमूने में अन्य हेजार्ड (खतरनाक) की उपस्थिति नहीं है। अतः व्यावहारिक रूप से इकट्ठा किए गए सूचनाओं के आधार पर एक HACCP फ्रेमवर्क विकसित किया गया जिसके इस्तेमाल से भारतीय श्रिंप फार्म को प्रतिजैविकी अवशेष से बचाया जा सकता है।

ABSTRACT

Aquaculture has developed to become one of the fastest growing food production systems in the world with a growth rate of 9.6% per year in the last decade and bulk of its output currently being produced within developing countries. India is the 5th top most cultured shrimp producer and farmed shrimp contributes about 60.5% by volume and 82% by value of total export. Shrimp farming has become an important component of the Indian aquaculture industry and about 75% of the farming area is culturing the species *Penaeus monodon*. With the establishment of WTO and its SPS agreements and increasing concern of health aspects, shrimp farming in India has severely been affected due to the rejection of several shrimp consignments by the major importing countries such as EU, USA and Japan, on the grounds of presences of traces of banned antibiotic residues. India being a member country of WTO and to promote the Indian shrimp products at international market, there is no option except to implement the international quality standards throughout the food chain. The Hazard Analysis Critical Control Point (HACCP) is a risk management program that has been accepted on a worldwide basis to prevent food safety hazards. In addition to food safety issues, HACCP principles provide disciplined scientifically based structures that can be used to develop risk management program addressing a variety of issues. Keeping in view of this, a HACCP study has been conducted during the year 2003-04 in 120 shrimp farms located in coastal districts of Indian states viz. Andhra Pradesh, Tamil Nadu, Gujarat, Maharashtra, Karnataka and Kerala to analyze the potential hazards and existing farming practices that are responsible for the use of antibiotics. Site selection, pond preparation, water stocking, shrimp seed, feed, water quality, application of antibiotics, farm management practices and harvesting were the identified nine Critical Control Points (CCP's) in the Indian shrimp farms. In the present study, none of the farmers except one, revealed the use of antibiotics. The results showed that 56.9% of east coast and 38.4% of west coast farmers were facing the problem of WSSV. Most of the farmers had the crop losses due to seed quality (23.3%), Climate (20.8%) and water quality (19.1%). Majority of Andhra Pradesh (13.3%) and Karnataka (53.8%) farmers were facing the problem of loose shell syndrome. Most of the farmers associated the good farm management practices with that of successful farming. Out of the total respondents, about 30% suggested that subsidies and lab facilities would bring good results in shrimp farming. There was negative correlation at 5% level of significance between the use of antibiotics and water source and positive correlation between the use of probiotics, education level, and water source at 1% level of significance. To fix the critical limits (CL's) for the identified CCP's and to apply control measures an experimental trial was conducted to estimate the withdrawal period for Oxytetracycline, a commonly used broad spectrum antibiotic, with *P. monodon* reared in FRP tanks. Samples were analyzed using HPLC using UV detector. The results showed that 21 days of withdrawal period is required for the complete elimination of any antibiotic in the body of shrimp. ANOVA results indicated that there was no significant variance between the control and treatment means of the observed F- value with respect to length-weight relationship. The microbiological analysis of commercial shrimp feeds of 10 different brands collected from the growers of Andhra Pradesh showed that the coliform counts were in the range of 1×10^2 to 4.2×10^2 cfu/g and total plate count ranges 1×10^1 to 3.2×10^3 cfu/g. The physical hazards revealed the poor texture of shrimp feeds and absence of other hazards in most of the feed samples. A HACCP framework was thus developed incorporating the field generated information using generic HACCP model that is applicable at Indian shrimp farms to prevent antibiotic residues

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Introduction

1. INTRODUCTION

Aquaculture industry, which is a global supplier of fish, crustaceans and molluscs, has developed to become the world's fastest growing food production system with a growth rate of 9.6% per year in the last decade. It increased from 3.9% of total production by weight in 1970 to 29.9% in 2002 (FAO, 2004), the major chunk of increase being in China, India and South-east Asia using proven sustainable systems. Asia with its 90% share in the world production is the main source with China in the lead contributing 57% of the world production followed by India with 9% of world production (FAO, 1997a&b). Among the various exportable fishery products, shrimp occupies the major share and its production through both capture and culture has expanded. Presently the highest shrimp producing country is China with 1.3 million mt of production per annum followed by Indonesia, Thailand and India (FAO, 2003, 2004).

India being endowed with a coastline of 8,119 km, a continental shelf of 0.5 million sq.km, an extensive Exclusive Economic Zone of 2.02 million sq.km, and about 1.24 million ha of brackishwater area offer an immense scope for development of fisheries and aquaculture. The fisheries sector including brackishwater aquaculture contributes around 1.5% to the total Gross Domestic Product (GDP) and around 5% in the GDP from the agriculture sector. Export of seafood, the majority of which is cultured shrimp, contributes over Rs.6, 000 crores. The annual growth in fisheries sector is 8.70% with marine and inland sectors contributing 2.10% and 6.60% respectively with an overall average of 4.35%. The contribution of Indian fisheries sector in the international trade is 1.1% of trade and 2.4% in value.

India is one of the largest producers of shrimps exports of which yielded Rs. 4,235 crores during 2001-02 and as much as 89.57% of it was contributed by aquaculture. In terms of quantity, aquaculture contributed 65.72 % of the total 1.23 lakh tonnes of shrimps exported by India in 2002-03. The major markets for Indian shrimp used to be mainly USA followed, in

decreasing order by Japan and the European Union, but during 2003-04 the trend changed with the EU occupying the 1st position followed by the USA and Japan. During the fiscal year 2004-05, India exported 4,61,329 tonnes of products fetching Rs.6, 646.69 crores. Frozen shrimp contributed around 1,38,085 tonnes worth of Rs.4, 220 crores and 63.50% in terms of value.

In India, shrimp farming has become immensely popular with about 1,57,000 ha under culture and an average annual production of 100,000 tonnes. About 91% of the shrimp growers have a holding in between 0 to 2 ha, 6% between 2 to 5 ha and the remaining 3% have an area of 5 ha and above indicating maximum contribution by the small farming system to the total cultured shrimp production in the country. Shrimp farming provides employment directly to about 0.3 million people and indirectly to 0.6 to 0.7 million people mainly in states bordering the east coast states of Andhra Pradesh, West Bengal, Tamil Nadu and Orissa since a decade. At present both area and production are the highest in Andhra Pradesh (79,760 ha; 51,230 mt) accounting for 50.57% and 50% of total cultured shrimp area and production, followed by West Bengal (47,650 ha, 26,800 mt) with 30.27% and 26% respectively. Kerala, Karnataka, Goa and Gujarat are the other states in the west coast where shrimp farming is being practiced (Cyriac, 2002).

In both Andhra Pradesh and Orissa, farmers owning less than 2 ha dominate (96.18% and 97.88% respectively), while in West Bengal and Tamil Nadu, only 38.15% and 16% of the farms respectively are less than 2 ha (AAI, 2001). Only about 10 out of 500 corporate sector companies participated in shrimp farming activities, mainly in Tamil Nadu, Andhra Pradesh, Orissa, Goa, Maharashtra and Gujarat. Farming of shrimp, mainly *Penaeus monodon*, has become an important component of the Indian aquaculture industry commencing the early 1980's and became a significant commercial activity by 1990, mainly due to the use of under-utilized water logged areas and adoption of improved scientific farming practices in various parts of the country.

Despite the progress, the shrimp industry has been facing severe crisis especially from 1994 owing to the outbreak of infectious diseases like White Spot Syndrome, which have caused mass mortalities and extensive damage

to the system. During 1999, the Hon'ble Supreme Court of India imposed a complete ban on all aquacultural activities within the Coastal Regulatory Zone.

More recently, India is facing a great problem regarding the use of antibiotics and chemicals in shrimp farming. Most of the people in the world especially developed countries have concern about consumption of shrimp with out antibiotic residues. Crop losses due to diseases during 1994-98 were estimated to be as high as Rs.1, 203 crores. Although a wide range of pathogens represented by viruses, bacteria, fungi and protozoans invade the shrimps in the culture system, the diseases caused by viruses have emerged as the most virulent. To control the disease outbreaks and secondary infections the shrimp growers resorted to use indiscriminate use of several chemicals, antibiotics and other drugs. Even as they failed in controlling the diseases, the indiscriminate use of antibiotics caused the problem of tissue residues in the harvested shrimps. The European Union (EU) rejected 12 shipments of shrimps involving 130 tonnes, during the year 2002 and placed 4 Indian processing plants under black list mainly due to the presence of antibiotic residues. Eighteen out of the 32 rejections in 2002, 18 were due to antibiotic residues (Ajayan, 2002).

The establishment of World Trade Organization (WTO) in January 1995 allowed the importing countries to impose strict quality standards. As India is one among the 148 member countries of WTO, it was compelled to implement the quality standards. Health safety and technical barriers to shrimp trade follows the Codex Alimentarius Commission's recommendation to adopt a food safety management system called 'HACCP'. These recommendations were endorsed and made virtually mandatory by the WTO in terms of Sanitary and Phytosanitary (SPS) and Technical Barriers to Trade (TBT) agreements. The SPS include those proposals that aim to protect human or animal health from food borne risks, and pests or diseases while TBT covers all technical regulations, voluntary standards and procedures including those on encompassing labeling requirements and regulations on nutritional claims. Thus, the post WTO era, shrimp farming in India faced a

serious setback due to the rejection of several Indian consignments based on antibiotic residues. Resultant economic losses were heavy and leading several shrimp farmers especially from Andhra Pradesh to commit suicide.

The indiscriminate use of antibiotics with improper labeling posed several hazards on planning Govt. agencies like Marine Products Export Development Agency (MPEDA) to implement the HACCP system throughout the food chain to control the food supply hazards. Increasing number of rejections of Indian seafood export consignments by the European Union (EU) forced the Export Inspection Agency (EIA) of India to ban production and export of some units. This jolted the Rs.15,000 crores seafood industry (Anon, 2002). MPEDA banned about 20 antibiotics including chloramphenicol, nitro furan etc in shrimp culture practices and fixed Maximum Residual Level's (MRL's) for other antibiotics. These controlling measures did not bring about any change in the situation and antibiotic residues were found frequently in the Indian consignments. The antibiotic residue has once again become a major issue with the recent rejection of shrimp consignments exported to Japan and fetched loss of Rs.20 crores. The shrimp exported from the state of Andhra Pradesh have been found to contain residues of Nitrofurans and its metabolites and the raw material had poor quality due to the muddy smell (Anon, 2005a; Anon, 2005b).

Quality is the most important factor for exporters in accessing the food markets of the developed world. In response to the requirement of importing countries, 380 Indian shrimp processing units have already volunteered to implement the HACCP concept. In order to compete in the world trade the option left is to implement the international quality standards the farm level itself with a 'farm to table' approach as in the case of exporting countries like Thailand, which have already started implementing the HACCP system at farm level.

To overcome the problem of antibiotic residues and to improve the quality of the raw material, it is essential to implement quality standards such as Hazard Analysis Critical Control Point (HACCP) throughout the food chain. There is scope for expansion of area under brackishwater culture as presently

only 10% of potential area is under culture. In order to ensure that shrimp cultured products exported from India are free from antibiotic residues, HACCP at farm level seems to be the most viable solution followed by Good Aquaculture Practices. Countries intending to export shrimp products to the EU, Japan and USA must follow the HACCP system regulation initiated by these countries.

Keeping in view of the existing scenario in the Indian shrimp industry, the present study had been carried out with the following objectives so as to develop a HACCP framework applicable at the growout level of Indian shrimp farms.

- To analyze the existing shrimp farming practices in the major shrimp farming states;
- To measure level of adoption of scientific farming practices;
- To identify the different sources of hazards leading to the presence of antibiotic residues;
- To estimate the hazardous substances entering through the feed; and
- To generate information in order to establish critical control points and their critical limits.



Review of literature

2. REVIEW OF LITERATURE

2.1 Global Aquaculture

The contribution of aquaculture to world food supply of aquatic products has been increasing over the past 10 years in comparison to fisheries, growing from 15 to 28% of total production between 1988 and 1997 (FAO, 1999). Shrimp, which does not naturally feed high in the trophic level but is mostly reared on artificial feed, has become a significant culture commodity. All regions except Africa and the countries of the former USSR, have recorded a significant increase in *per capita* production commencing 1984 (FAO, 1999). The contribution of aquaculture to the global aquatic food supplies has increased steadily over a period of about 15 years by comparison to the capture fisheries; the share of aquaculture in the total supply has grown from 12 to 28%, tantamount to the position that nearly every third kg being consumed is cultured (DeSilva, 2001).

The global shrimp farming industry had a rapid growth in the 1980s mainly due to technological breakthrough such as hatchery and feed, high demand for shrimp resulting in high price and high profit of shrimp farming and public support. However, the growth has slowed down since 1991 due to serious outbreaks of shrimp diseases that reported in most of the major producing countries. Viral diseases have reduced shrimp production farmed shrimp accounted 27% of total shrimp production from wild-caught and farm-raised sources; Asia produced about 78% of farmed shrimp and western countries 22% (Shang *et al.*, 1998; Datta.2001).

Some 63 countries were listed as having produced shrimp at one time or another from 1984 but by 1994 only 29 countries were found in all regions, including Europe and the Middle East, to be adopting shrimp culture (Srisomboon and Poomchatra, 1995). Many of the countries in the Middle East region including UAE, Kuwait and Yemen have initiated move to venture into shrimp farming. Extensive and intensive methods of culture are practiced

in Ecuador and all the Latin American countries from Mexico to Peru produce shrimp (FAO, 1999; Yap, 2000).

Marine shrimp culture dominates crustacean culture representing 96% that is done in brackishwater and 73% of all crustacean aquaculture. The tiger prawn, *Penaeus monodon*, contributes in excess of 50% to the total followed by white leg shrimp, *P. vannamei* (18%) and the oriental or fleshy prawn, *P. chinensis* (10%) (Shang *et al.*, 1998; FAO,1999).

2.2 Shrimp Aquaculture in India

About 85 species of shrimps are known to exist in Indian waters, of which 55 species are reported (Yadava, 2002) either as commercially important or having considerable demand in the local as well as international market. Among these, *Penaeus monodon*, *P. indicus*, and *P. merguensis* are having high demand and are candidate species for cultivation. The black tiger shrimp *Penaeus monodon*, however, is the mainstay of the industry. Shrimp farming, which was traditionally practiced in the coastal states of West Bengal and Kerala, is now common in not only these two states, but also in all the other coastal states, *viz.*, Orissa, Andhra Pradesh, Tamil Nadu, Karnataka, Maharashtra, Goa, and Gujarat.

About 1.2 million ha of potential area suitable for shrimp farming have been identified in India; of this, the area actually under cultivation was 65,000 ha in 1990 but increased to slightly more than 1,57,000 ha by 2002 (Yadava, 2002; Ravindranath and Madhavi, 2004). The number of shrimp farms in India was estimated to be nearly 93,000 with an annual production around 1,00,000 tons in 2002 (Yadava, 2002).

In India, shrimps available were locally consumed till 1950's and thereafter, until late 1980's, exported in dried form to East Asian countries like Hong Kong, Singapore, Myanmar and Sri Lanka; supply was, however, mainly from capture component only. By 1990's the contribution of culture sector to overall shrimp production became noticeable, more so with the trend of no growth in capture shrimp sector. Eventually, as at present, the culture shrimp segment has come to occupy around 50 % of the total shrimp

catches. It can be said that culture shrimp production has become a savior to maintain and enhance Indian shrimp exports level (Anon, 2001).

Improved means of preservation, namely freezing and canning, added value to the frozen product over the dried product. The first consignment of frozen shrimp valued at Rs. 60 lakhs was exported in 1952; ever since, the export of marine products has grown to levels whereby shrimp contributes about 35% by volume and around 70% by value to the marine exports (MPEDA, 1996). The major markets for Indian shrimp are Japan, Western Europe and USA.

The growth of shrimp farming was slow in the decade of eighties and an exponential growth in farming took place during early nineties due to the attractive monetary gains, improved scientific farming high export demands, liberalized economic policies of the government, and higher participation of farmers, entrepreneurs, and the corporate sector, resulting in increase in area by 78% and in production by 195% (Vasudevappa and Seenappa, 2002). Production and productivity trends indicate gradual increased from 1990 to register an all time record production of 0.103 million MT, 0.66 million MT/ha/yr in 2002 (Cyriac, 2002).

Shrimp culture in India is practiced in four types, *viz.* traditional culture, extensive, modified extensive culture and semi-intensive culture (MPEDA, 1996b). Semi-intensive and intensive shrimp farming systems are characterized by high inputs of fertilizers and supplementary feeds and increased load of nutrients, organic matter and other wastes that can affect water quality in ponds and receiving waters.

About 91% of the shrimp farmers in the country belong to the medium (2-5 ha) category and the rest are large farmers (Yadava, 2002; MPEDA, 1996; Anon, 2003a). The state of Andhra Pradesh continues to lead in shrimp and prawn aquaculture in India. There are around 73,000 aqua farmers in the state of Andhra Pradesh. The country has about 92,591 farms of which 90.57% are less than 2 ha size, 5.80% are 2 to 5 ha size, 2.13% are 5 to 10 ha and farms of more than 10 ha size account for only 1.54%; this indicates

the maximum contribution of small farming system to the total cultured shrimp production in the country. Andhra Pradesh accounts for 50-57% and of total shrimp culture area and 50% of production followed by West Bengal, Kerala, Karnataka, Gujarat and other states where shrimp farming is being practiced (AAI, 2001; Cyriac, 2002).

Shrimp farming in most of the states are creek based and are of improved traditional type. In Andhra Pradesh creek based farming represents 96%. In Orissa 45% of the shrimp farms are at the Chilka lake periphery and 55% is creek based while in Tamil Nadu seawater-based farming represents 44.36% (AAI, 2001). It was mainly represented by extensive (70%), semi-intensive (25%) and intensive systems (5%) during the boom period (Rosenberry, 1995). About 10 corporate sector companies out of 500 companies participated in the farming activity mainly in Tamil Nadu, Andhra Pradesh, Orissa, Goa, Maharashtra and Gujarat (Anon, 1999).

Shrimp culture in India is mainly an enterprise of small and marginal farmers and has created direct employment to more than 0.3 million people and indirect employment to over 0.7 million people (Vasudevappa and Seenappa, 2002). Shrimp farming has brought in several support services under backward and forward linked sectors assisting development of the farming in the country. Infrastructural improvements *viz.* roads electricity, sanitation and housing in villages (CIBA, 1995), additional facilities such as telephone, transport, communication linkages and markets (Rajalakshmi, 2001), allied sectors *viz.* brood stock and spawner supply, supply of equipments and other material for hatcheries and farms, supply of feed manufacturing units, feed ingredients, feed supply, ice plants, processing plants, etc. are some of the major one's linked to the farming in the country (Vasudevappa and Seenappa, 2002; Alagaraswami, 1995).

2.3 Issues in world shrimp farming

Non-availability of quality shrimp seed from hatcheries, quality feed, mismanagement of water and soil quality, feeding, lack of technical assistance for diseases diagnostics, fluctuating farm gate prices, problems in

processing and international market demand and constraint of the individual investors (capital, land, water and management skill) are recognized as the major factors governing the choice of farming system and success of farming (Krishnan *et al.*, 2001; Vasudevappa and Seenappa, 2002; Chamberlain, 2003a).

Boyd and Tucker (1998) reported that water quality problems have affected about 33% of shrimp farms in the USA and that the major problems were excursion of salinity outside of the desirable range, high turbidity and plankton blooms. Sustainability and environmental degradation coupled with numerous viral and other diseases are the biggest problems in the shrimp culture (Treece, 2000).

Shrimp farmers worldwide struggled with trade issues, new antibiotic standards and a glut of farmed shrimp. Shrimp farmers in as many as 12 countries faced anti dumping charges brought by shrimp fishermen in the South-eastern United States. Shrimp importing nations have set the tolerance for antibiotics in farmed shrimp at a level close to zero (0.3 ppb, with the ability to detect down to 0.1 ppb). Shrimp farmers have no choice; they either stop using antibiotics or their products will be rejected or in some cases destroyed by the importing countries (Rosenberry, 2003).

Sustainability and environmental degradation coupled with numerous viruses and other diseases are the biggest problem in the shrimp culture (Banora, 1999; Treece, 2000; Bhatta, 2004). Shrimp diseases and poor water quality were the chief reasons that world shrimp production declined slightly from 1995 through 1997; the industry appeared to be rebounding from then problems and was producing more shrimp than ever before in 1998. However, the WSSV problem continued to plague the industry on a worldwide basis. China's shrimp-farming industry boomed until the early 1990s, but then crashed. India and Indonesia boomed until the mid 1990s and then leveled off or declined when problems related to over expansion took place. Honduras, Mexico and Columbia all had substantial industries at one time, but production declined with diseases, water quality and other problems.

International disputes over seafood safety have affected trade opportunities for producers, exporters and importers. In 1994, the Spanish government rejected two shipments of squid from the United States (USFDA, 1991). In 1997, the European Commission (EC) banned shrimp imports from Bangladesh because processing plants in Bangladesh did not meet EC standards.

2.3.1 Use of Chemicals, Drugs and Antibiotics

Animal drugs may be used in the raising of aquatic species: (1) to treat or prevent disease, (2) to control parasites, (3) to affect reproduction and (4) to tranquilize. Illegal residues of drugs may occur in aquaculture species because of the use of unapproved drugs, use of drugs not in accordance with the approved labeling directions, failure to follow approved withdrawal times, or use of general purpose chemicals not labeled or approved for drug use. There are only a few approved drugs for aquatic species in the USA and approval of the FDA is required before any animal drug is used to ensure that unsafe drug residues will not occur in edible tissues when animals are treated following approved label directions (Anon., 2001; Johnson and Santillo, 2002).

Greslund *et al.* (2003) found that marine and brackishwater shrimp farmers in Thailand used on average of 13 different chemicals and biological products as also soil and water treatment products, pesticides and disinfectants. Primavera *et al.* (1993) reported about a similar situation in shrimp farms in the Philippines. Angelillo *et al.*, (2000) conducted a study to evaluate knowledge, attitude and behavior concerning food borne diseases and food safety issues among food handlers in Italy, and strongly emphasized the need for the educational program for improving knowledge and control food borne diseases.

As in other animal production sectors, antibiotics are used in aquaculture during both production and processing, mainly to prevent (prophylactic use) and treat (therapeutic use) bacterial diseases (FAO, 1997; FAO, 2002; Griffith, 1999). Antimicrobial substances are often used not only

as prophylactics, but as growth promoters (Willis *et al.*, 1999; Bimal, 2000), but the nature of growth promoting mechanism of antibiotics is still not clear (Ghosh and Ray 2003). The observed growth promotion may be due to the fact that antibiotics control the level of infection in animals, or the antibiotics might have more specific effects in promoting the metabolism of animals (Lalumera *et al.*, 2004).

In aquaculture, antibiotics are generally administered in feeds (Srisomboon and Poomchatra, 1995), having been either added during feed manufacture or surface-coated on to pellets by the manufacturer or the farmer. During outbreaks of disease, farmers may apply antibiotics using other routes (FAO, 2004). Feed is the most important input contributing to the waste although fertilizers, chemicals, antibiotics and drugs may also contribute. Medicated feed is usually prepared on site by mixing the antibiotic with pelleted feed and surface coating with an agent such as oil, gelatin or whole egg or simply mixing with trash fish (Inglis, 2000; Sinhaseni *et al.*, 2000).

Reilly and Kaferstein (1997) reported on the uncontrolled use of antibiotics as therapeutic agents, growth promoters and for increasing the efficiency of feed utilization in semi-intensive and intensive aquaculture systems. Antimicrobial substances are however often used not only for disease-prevention, but as growth promoters during the production of animals for the human consumption (Willis *et al.*, 1999).

2.4 Implications of the use of antibiotics in aquaculture

The use of antibiotics in shrimp aquaculture practices will lead to development of drug resistant bacteria, retention of drug residue is farmed shrimp, post-health hazard to the consumers. In the shrimp farms on an average of 60 mg/m² of waste is produced for every kilogram of shrimp produced. The use of antibiotics/antibacterial chemicals in the farm either as therapeutic, prophylactic or growth promoters, destroys the environmental micro flora that are useful in mineralization of the waste with their scavenging action. Indiscriminate use results in development of drug resistant bacteria,

some of which are human pathogens. 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 (Surendran, 2003).

Anesthetics, disinfectants and antibiotics would cause lethal or sub lethal effects on non-target organisms and might lead to development of drug resistance in pathogen (Gupta, 2001). Antibiotic use increases the risk of contamination, as more fish or crustaceans are exposed to sub therapeutic levels of antibiotics in feeds to prevent or reduce the incidence of infectious diseases. Dosage rates, frequency, and with holding or washout times prior to harvest are critical determinants in assuring that antibiotic residues are not present in final aquaculture products (Ruth, 2002). Residues of antibiotics caused by irresponsible sales and abuse of veterinary medicines in intensive and semi-intensive aquaculture systems create a first rate problem of food security (Reilly and Kaferstein, 1997).

2.4.1 Antibacterial Resistance

Lee and Edlin (1985) suggested that tetracycline resistance has been evolving for a very long time (millions of years), perhaps in response to competition with organisms producing tetracycline like substances. Laboratory studies have shown that expression of resistance is selected against in the absence of the drug (Chopra, 1985; Lee and Edlin, 1985; Hamilton-Miller, 1990; Modi *et al.*, 1991; Smith *et al.*, 1994; Inglis, 2000). Anesthetics, disinfectants and antibiotics would cause lethal or sub lethal effects on non-target organisms and might lead to development of drug resistance in pathogens (Gupta, 2001).

With some drugs (e.g. Oxytetracycline), many groups of bacteria display a clear bimodal distribution of sensitivity, and classification into sensitive or resistant is easy. Problems arise with strains designated of intermediate sensitivity as happens when resistance is increased in small steps (Inglis *et al.*, 1991). Hamilton-Miller (1990) attributed that the anti-microbial resistance with frequency of use in an environment. Prescott and

Baggot (1988) reported on the resistance to penicillin of *Staphylococcus aureus* and resistance of *Escherichia coli* from pigs to sulphanamides, streptomycin and tetracycline. Bacterial strains resistant to quinolone and oxolonic acid were isolated from fish by Hastings and McKay (1987) while Inglis *et al.* (1991) reported that 40-50% of isolates of *Aeromonas salmonicida* in Scotland were resistant to many antibiotics.

2.4.2 Antibiotic Residues in Aquaculture Products

The presence of residual antibiotics in sediments and in the tissues of shrimp may have public health implications, and then substances are regarded as dangerous for humans and preventive measure suggested to ensure optimal diet quality (Boonyaratapalin, 2000). The European Union and the USFDA (USFDA, 2001) as also Japan have notified that residues of the antibiotics listed in their notification should not be present in the imported shrimp. Antibiotic residues have been detected in a small portion (8-9%) of tiger prawn tested in the UK (Willis *et al.*, 1999). Surendran (2003) also reported that chloramphenicol and nitrofurans were detected in Indian shrimp exported to Spain, Netherlands and UK in ppb levels. All the consignments were confiscated and destroyed and fetching loss of nearly Rs.100 crores. 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 study conducted by CIFT, Cochin showed that out of 2086 samples of farmed shrimp (tiger prawn, white prawn and the freshwater prawn or scampi) tetracyclines were detected in 13 and samples and chloramphenicol in 28 samples. During the year 2002, the detection of chloramphenicol in internationally traded shrimp products has caused much concern. The substance has been found in cultured products, resulting in a slow down in imports, causing economic loss among the concerned producers and reflecting negatively on all shrimps and on aquaculture overall (FAO, 2002).

2.5 Oxytetracycline (OTC) in aquaculture

The tetracyclines were introduced in the 1950 (Snieszko *et al.*, 1951) and found to be highly effective against Gram-negative pathogens. Oxytetracycline soon was widely used to treat many bacterial diseases in many spheres of aquaculture in Europe, the USA and Asia but by 1990 countries varied widely in the drugs they used in their aquaculture systems (Schnick, 1992; Alderman and Micheal, 1992; Schlotofeldt, 1992). Alderman (1992) reviewed the main groups of drugs used in aquaculture, their application, toxicity, pharmacokinetics and residues and provided a good background against which subsequent developments should be assessed. Oxytetracycline is a broad-spectrum antibiotic used to control enteric and systemic vibriosis through medicated feed. In the case of giant freshwater prawn, Oxytetracycline is used in the treatment and control of infectious bacterial diseases such as vibriosis and is generally given by mixing with prepared diets, more often as formulated medicated diet at the concentration rang 2.5 g/kg diet *ad libitum* for 5 days.

Studies have shown that bioavailability of oxytetracycline is very low, being 0.38% for carp and 1.26% for trout after a single oral dose of 60 mg/kg, while plasma concentrations of 0.65 and 0.37 µg/mg were achieved in trout at 10°C and 19°C and 0.15 and 0.81 µg/m; in carp at 8°C and 20°C respectively (Nouws *et al.*, 1992).

Absorption of oxytetracycline from the intestine of fish has been shown to be poor (Cravedi *et al.*, 1987) being 7.1% compared with 38.1% for oxolonic acid. Maximum levels achieved in muscle tissues are low, 0.6-1.6 mg/ml (Inglis, 1999). It was estimated that only 20-30% reached the fish and the rest entered the environment mostly bound to sediment particles (Jacobson, 1989). Oxytetracycline from fish or fish feed might then be dissolved in the water or deposited in sediment below the cage. Factors which affect the concentration of antibacterial drugs in marine sediments include the amount used, currents at the time of medication and temperature (Jacobson and Berglind, 1988). Several authors reported on the pharmacokinetics and

bioavailability of oxytetracycline, and withdrawal times in rainbow trout (Abedini *et al.*, 1998; Jacobson, 1989; Rogstad *et al.*, 1991).

The only Codex Alimentarius Commission (CAC) MRLs for aquaculture species listed in the database are for the administration at 100 µg/kg to 'fish' and 'giant prawn', but several additional MRL proposals from 'Joint FAO/WHO Expert Committee on Food Additives' (JECIA) are now within CAC system (FAO, 2004).

Once in solution, oxytetracycline is likely to very quickly be diluted to negligible concentrations and then inactivated. Tetracycline in aqueous solutions is degraded, by photo decompositions and half-life is dependent on temperature, pH, air saturation and light density (Smith *et al.*, 1944; Oka *et al.*, 1985; Samuelsen, 1992). Bjorklund *et al.* (1991) conducted a study at two separate farms, where the half-lives of oxytetracycline in sediments were found to be 9 days and 60 weeks. Oxytetracycline may be lost from sediments by leaching, degradation or formation of stable complexes (Inglis, 1990). Touraki *et al.* (1954) reported that 70-80% of the orally administered OTC remains in the environment and there is a high persistence of antibiotics in sediments in fish farms. Williams *et al.* (2002) found that under recirculating system conditions, the OTC concentration increased in trout during the 10 days of treatment, declined after the medicated feed was stopped and was well below 2 µg/g by the end of the 21 days withdrawal period. Lalumera *et al.*, (2004) reported that oxytetracycline and flumequine were detected in the sample sediments of trout farms and seabass farms.

Oxytetracycline is widely used in the treatment of bacterial infections in aquaculture. It has been reported to have immunosuppressive effects and results from a histological study indicated that it might cause liver damage. Furthermore, bacterial resistance has been frequently observed in diseased salmon and shrimp. The elimination half life ($t_{1/2}$) of OTC was 16.12 hours and 90.3 hours respectively was reported in *Penaeus chinensis* (Weifen *et al.*, 2004).

A withdrawal period of 4 weeks before marketing was recommended for oxytetracycline (Shankar and Mohan, 2002). The recommended withdrawal period by MPEDA (1996) for oxytetracycline in different rearing water temperatures were 60 days (less than 12°C), 40 days (12-22°C) and 15 days (> 22°C).

Ueno *et al.* (1999) reported that the average recoveries and coefficient of variation for oxytetracycline were 97 and 1.3% with detection limit of 0.02 µg/g in muscle tissue of shrimp, by using HPLC analysis.

In an experiment conducted by Selvin and Lipton (2004) reported that *Penaeus monodon* fed for 7 days with 50-100 mg/kg shrimp of OTC were examined for residual accumulation and depletion. A residue of 3.47 µg/g shrimp tissue was found after 1 day of post dosing and a substantial quantity of residue 1.56 µg/g was observed upto 15 days of post-treatment. By the 20th day of experiment, the OTC concentration was found in trace quantity 0.42 µg/g or below detectable limit. In a study, when food containing oxytetracycline was discontinued, retention of this antibiotic in the white shrimp *Penaeus setiferus* was undetectable within 3 days for the 1,000 mg group and within 2 weeks for the groups on 5,000 and 10,000 mg/kg oxytetracycline diets (Corlis, 1979).

Bebak-williams *et al.* (2002) assayed oxytetracycline concentration in medicated feed and unmediated feed within 2 weeks of the manufacture data and reported 6.2 g of OTC-HCl/kg and 6.0 g of OTC-HCl/kg of two sub-samples of medicated feed. For unmediated feed OTC was measured at 21 and 18 mg/kg. In other trial samples were analysed about 3 months after the manufacture data. At that time OTC was measured in medicated feed at 6.2 and 5.5 g OTC-HCl/kg and it was <10 mg/kg in unmediated feed for both samples. The medicated feed was added 6.6 g OTC-HCl/kg with 6.1 g/kg of OTC concentration. The potential presence of two common antibacterial agents, Oxytetracycline (OTC) and Oxolinic acid (OA) in Mediterranean fish farms was estimated from the measurements of these drugs in the faecal excretions of gill thread sea bream (*Sparus aurata*) and sharp snout sea bream (*Diplodus puntazzo*). Oxoline acid was found to be well absorbed by

gilthead sea bream (92%) and sharp snout sea bream (88%) while the absorption of OTC was found to be considerably lower in both species (27 and 40% respectively). Further, drug may also be released via un eaten medicated feed, leached drugs and other routes of fish elimination such as renal excretion and bran dial excretion (Rignos *et al.*, 2004). Oxytetracycline is approved for use in aquaculture in the USA, Canada, European countries and Japan, whereas, its usage is not approved in Australia (Abraham, 2002).

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Onkong *et al.* (2000) reported that under practical conditions where farmers use oxytetracycline at the rate of 5 g/kg of shrimp feed, the longest elimination of the drug from any tissue was 18 days, and suggested that the withdrawal period between the use of OTC and harvesting should be no less than 21 days.

In an experiment conducted by Selvin and Lipton (2004) reported that *Penaeus monodon* fed for 7 days with 50-100 mg/kg shrimp of OTC were examined for residual accumulation and depletion. A residue of 3.47 µg/g shrimp tissue was found after 1 day of post dosing and a substantial quantity of residue 1.56 µg/g was observed up to 15 days of post-treatment. By the 20th day of experiment, the OTC concentration was found in trace quantity 0.42 µg/g or below detectable limit. In a study, when food containing oxytetracyclin was discontinued, retention of this antibiotic in the white shrimp *Penaeus setiferus* was undetectable within 3 days for the 1,000 mg group and within 2 weeks for the groups on 5,000 and 10,000 mg/kg oxytetracycline diets (Corliss, 1979).

2.6 Food Safety Issues and Public health Concerns

USDA predicts radical changes in the next 20 years in food purchase and eating habits, with more health conscious consumers, and increased immigration from East and Southeast Asia, resulting is a 25% increase in seafood purchase (Anon., 2003b). As the world food supply becomes more global, food quality and safety issues will become more prominent. Many developing nations have realized that one of their greatest natural resources is food, which can be traded with developed nations (Suwanrangsi *et al.*, 1999).

Chemical contamination can result from local spills or dumping of pesticides, industrial chemicals, heavy metals, and petroleum products. Different countries allow the use of different vaccines, feed additives and antibiotics for farm raised fish and fishery products and therefore, in some cases, residues from these production inputs may raise food safety concerns (FAO, 2001a). Residues may remain in fish used for human consumption and antibiotics released into the environment can lead to the development of antibiotic resistant bacteria in the food chain (Reilly and Kaferstein, 1997; Hagler, 1997).

Food borne diseases most seriously affect children, pregnant women, the elderly and people already affected by other diseases. Food borne

diseases not only significantly affect people's health and well-being, but they also have economic consequences for individuals, families, communities, business and countries (WHO, 2002; World Bank 1997; World Resources Institute, 1998). For example, 40 million people become infected each year from trematode parasites by consuming raw or inadequately processed shellfish, freshwater fish, and aquatic plants (WHO, 1995).

A food safety survey by the USFDA in 1998 revealed that 12% adults ate raw oysters (Fein and Riggins, 1998). Most seafood associated illness reported by U.S. consumers point to consumption of raw bivalve mollusks and to unspecified and unknown food borne illness with Norwalk – like viral gastroenteritis symptoms (Ahmed, 1991). According to the U.S., Center for Disease Control and Prevention (CDC) surveillance data for food borne disease outbreaks indicates that 6.8% of 2,750 outbreaks during 1993-97 were attributed to consumption of shell fish and other fish (Olsen *et al.*, 2000).

The development of antibiotic resistance by pathogenic bacteria is considered to be one of the most serious risks to human health at the global level (Cahill *et al.*, 1996) because antibiotic resistance from bacteria associated with animals has been found to be transferred to human pathogens (Moriarty, 2001). Antibiotic residues in food can be dangerous to the consumer. Chloramphenicol is known to cause haemotoxic side effects particularly the chloramphenicol induced aplastic anemia, 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 and hence they are declared as zero tolerant antibiotics (Inglis, 2000).

There is a growing public concern about risks to consumers ingesting drug residues (Inglis *et al.*, 1993, Reilly *et al.*, 1997) and about effects on the environment. Many countries, however, do have national sampling and surveillance schemes in place to monitor veterinary drug residues in meat intended for human consumption and these include the products of aquaculture (Inglis, 1999). Drug excretion is usually described in terms of degree days (the product of temperature in degrees Celsius and time in days).

Show excretion provides an opportunity for the selection of resistant bacteria among the normal gut flora and in pathogens persisting in the host. This is a potential hazard in cold water aquaculture and this risk may be only minimal in warm water systems (Inglis, 1999).

2.5.1 Antibiotic Residues in Aquaculture Products

Antibiotic residues can occur in products when correct withdrawal times are not followed or when antibiotic sale and use are not controlled (Reilly and Kaferstein, 1997). Where antibiotics are incorrectly used or withdrawal times are not observed, residues in the fish flesh may pose a potential threat to consumers, especially those who are hypersensitive, leading to possible allergic reactions. Safe levels of residues for some of the more common antibiotics used in aquaculture have been recommended by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1996).

European Union had rejected several shrimp products mainly due to the presence of antibiotics and Japan has also been rejecting the products based on muddy smell and USA imposed antidumping due to the availability of Indian shrimp lesser than the rate of USA domestic market (Anon., 2004).

EU suspends shrimp and prawn imports (and other products of animal origin) from China because of residues from a banned antibiotic, chloramphenicol, and because of general deficiencies in the Chinese residue control system (GOI, 2002). The FDA response was to step surveillance for chloramphenicol residues and residues of other unapproved aquaculture drugs in shrimp and crayfish imports from all countries and to modify its testing methods so as to be able to detect the antibiotic at 0.3 ppb, equal to that of Canada and the EU; products with detectable levels of chloramphenicol will be detained and refused entry into the United States (US FDA, 2001).

Saitanu and Amornsinsin (1994) reported that tiger shrimp samples in Thailand revealed the presence of oxolonic acid and oxytetracycline were present in 12 and 9 samples respectively out of 21 positive samples of antibiotics.

In a study Mohny *et al.* (1997), juvenile shrimp fed for 14 days on oxytetracycline, medicated feed with a potency of 1500 mg of OTC kg⁻¹ of feed reached a level of 3.3 µg of OTC g⁻¹ of shrimp tail muscle within 4 days. By the fifth day following cessation of feeding medicated feed the drug content in shrimp tail muscle was less than the detectable limit for the method, or 0.4 µg of OTC g⁻¹ shrimp tissue.

International Trade - WTO

It is estimated that India's export earning through seafood products alone is approximately Rs. 6,300 crores. This market has been rapidly growing. India is signatory to the WTO, SPS and TBT agreements. With about 148 nations being a part of WTO, basic food safety standard as per Codex Alimentarius becomes mandatory. The SPS agreement covers all food hygiene measures and food safety measures such as the control of residues of veterinary drugs, pesticides and other chemicals used in meat production. The export of India seafood products would be affected by the insistence of USA and other developed countries on imports of food products only from those suppliers who established HACCP system at their plants (Ayyappan and Venkateswarlu , 2002; Kumar *et al.* 2004).

The United Nations World Health Organization (WHO) and the Food and Agricultural Organization of the U.N. (FAO) is backing HACCP since they recognize it is the best system available, and one that has been incorporated into the WTO (World Trade Organization) agreements controlling world trade. It is the reference standard for international trade i.e. no HACCP means no permit to import food into most western countries. It was also supported by many scientific organizations (e.g. the U.S. National Academy of Sciences) in the US and elsewhere. While there is no common agreement on food or fish safety at the international level, agreements exist on food trade that have implications for food safety and quality matters. The important one regarding food safety is the agreement on the application of Sanitary and Phyto-sanitary Measures (SPS), which introduced harmonization of SPS measures on as wide a basis as possible and member countries should base their SPS measures on international standards, where they exist. The same agreement

established that, regarding food safety, international standards, guidelines, and recommendations to be taken as reference will be those established by the Codex Alimentarius Commission (CAC) relating to food additives, veterinary drugs and pesticide residues, contaminants, methods of analysis and sampling and guidelines of hygiene practice (Josapeit *et al.*, 2001).

The principles of achieving harmonization of standards and equivalency in food control system and the use of scientifically based standards are embodied in the two binding agreement of the WTO. The SPS agreement confirms the right of WTO member countries to apply measures necessary to protect human, animal and plant life and health. The TBT agreement basically provides that all technical standards and regulations must have legitimate purpose and that the impact or cost of implementing the standard must be proportional to the purpose of the standard. This agreement also places emphasis on international standard (Valdimarsson *et al.*, 2004). WHO/FAO Codex Alimentarius incorporated HACCP in its general guidelines in 1997 and thereby created a starting reference for trade disputes under the WTO agreement on Sanitary and Phyto-sanitary measures (FAO, 2000).

Measures to Control Antibiotics Residues

Risk assessment is the tool used to evaluate the safety of food and food additives. Joint FAO/WHO Export Committee on Additive (JECFA) safety evaluation of food additives is a two-stage process. In the first stage relevant data are collected, including results of studies on experimental animals and where possible, human observations, including epidemiology studies. In the second stage data are assessed to determine whether a substance is acceptable for its intended use as food additive. This scientific process determines the possible adverse effects in humans resulting from exposure to a substance (Henry, 1997, Seni *et al.*, 2000).

The major shrimp importing countries have taken several measures to control the antibiotic residues in the imported shrimp. The EU enlisted nearly 300 pharmacologically active substances/ antibacterial / antibiotics / sulphenamids, which are either declared zero tolerant or for which maximum

residue limits (MRL) have been fixed. The acceptable levels of drug residues in the fishery products are based on the maximum residue limit (MRL). The MRL's are less than 50 ppb in most of the cases, viz. Chloramphenicol, Nitrofurantoin, Nitrofurazone, Nitrofurantoin, Furazolidone, Furaltadone and their metabolites (EEC, 2002).

USFDA (2001) imposed MRL's for seafood exported to USA; Oxytetracycline (2ppm), Sulfamerazine (Nil), Sulphadimethoxine / Ormetoprim combination (0.1ppm) and prohibited other antibiotics. Japan permits no antibiotic residue except tetracyclines, to maximum of 0.1 ppm in seafood. Inglis (1999) suggested that there is a real requirement for further study on the actual doses reaching fish in field conditions on the effects of leaching losses and feeding behaviour.

The application of probiotics as a microbial biotechnology to enhance and maintain aquacultural production levels is now gaining acceptance as a better, cheaper and more effective remedy than antibiotic use since probiotics promote shrimp health as well as consumer food and environmental safety to promote overall sustainable shrimp production. (Green and Green, 2003). Environmental certification systems are being developed to assure consumers that aquaculture products have been raised in a responsible manner (Chamberlain, 2003).

Antibiotic residues in shrimp not only had impacts on Japanese consumers and Thai economy but also to the American health authority (Srisomboon and Poomchatra 1995). Greslung and Bengtsson (2001) concluded in relation to shrimp aquaculture that 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.

In the WTO regime, as Indian trading partners are installing regulatory import controls, Export Inspection Council (EIC) of India has restricted its role to introduction of voluntary certification programs besides regulatory export control, especially in food sector, and is seeking recognition for EIC's

certification by official import control agencies of the trading partners as per provisions of WTO agreements to facilitate easier access to their markets for Indian exporters. In connection with this, EIC had issued notification dated 10th July, 2002 replaced the notification dated 17th August, 2001, whereby use of 20 antibiotics was banned and maximum residual limits were specified in respect of some other antibiotics, pesticides and heavy metals in all stages of handling in hatcheries, feed manufacturing farms, pre-processing and processing of the fish and fishery products in tune with the latest requirements of the EU, USA and Japan (Anon., 2004a).

There are 9 substances included in Annexure-IV of Regulation 237 EEC that may not be used in aquatic animal products; Chloramphenicol, Chloroform, Colchicines, Dapsone, Dimetridazole, Metronidazole, Nitrofurans (including Furazolidone) and Ronidazole. The presence of an Annexure-IV substance residue (including metabolites) is *prima facie* evidence of the use of prohibited substance in a food animal species. There are also provisions regarding the use of antibiotics to deal with emergencies (e.g. epidemics) and research. In general, banned antibiotics and banned veterinary drugs pose significant demonstrable risks to human health. Processing, cooking and frozen storage can reduce the residual levels of antibiotics (Lan *et al.*, 2001). However, data regarding the effect of processing, cooking and freezing aquatic animal products are scanty.

There are several possible strategies for limiting the commercial availability of antibiotics. The two most basic are, identifying the permitted antibiotics (and their MRLs) and prohibiting all others, or identifying the prohibited antibiotics and permitting all others. The first strategy is clearly more in line with the precautionary approach (FAO, 2002).

Antibacterial agents are widely used in aquaculture, and this is likely to remain. So, Inglis (2000) suggested that a code of practice be developed and supported for implementation in the future to ensure the greatest benefits in efficacy and commercial terms with minimal environmental impact and damage to health. Use of natural, herbal and homeopathic remedies and practices should be encouraged. Manufacturers should be required to provide

labels with the composition of chemical products, permissible uses, methods of application, environmental hazards and restrictions, storage, disposal and environmental and human safety precautions (Anon., 2003c). In view of the marketing problems associated with antibiotic residues in the shrimp tissues, it is prudent for the aquaculture managers to refrain from using antibiotics until the scientific basis of their usage and withdrawal periods are available (Abraham, 2002), all the more because the drug approval process and regulations differ in developed countries like USA, Canada, Australia, Japan and European Union. Diwan and Ayyappan (2004) suggested organic farming to overcome these problems and to produce fish and fish products through organic methods.

India has taken the antibiotic issue very seriously and has taken stringent actions that include legal notifications, campaigns in farms and hatcheries, and quality assurance inspections. Government of India issued notification in August, 2001, the Marine Products Export Development Authority (MPEDA) along with Export Inspection Council of India (EIC), the Ministry of Agriculture and the State Governments concerned under took a series of campaigns in the aquaculture farms in the country. MPEDA and other agencies directed the farmers, the hatchery owners and the processing units, feed manufacturers and manufacturers of various inputs for aquaculture to completely avoid the use of any of banned chemicals in their operations. EIC introduced a 3-tier monitoring system for surveillance of fish and fishery product units comprising of periodic monitoring visits, supervisory visits and annual corporate audit. The Government has introduced a series of hi-tech laboratories in major centers. As the International markets are taking steps to increase the traceability of the products to individual farms and supplier, MPEDA is making efforts to develop eco-labelling and organic shrimps (Mohamed, 2003).

The Seafood Exporter's Association of India has decided to purchase of raw material only if it is accompanied by a certificate (as required by the importing countries) of non-usage of antibiotics and antibiotic free product

from MPEDA/EIA/CIFT or any other Government approved laboratory (Balaji, 2002).

Withdrawal Period – Control related to the protection of public health

Good Practice in use of Veterinary Drugs (GPVD) as defined by the Codex Alimentarius Commission, is official recommended or authorized usage including withdrawal periods, approved by national authorities of veterinary drugs under practical conditions. The maximum residue limit for veterinary drug (MRLVD) is based on the type and amount of residue considered to be without toxicological hazard for human health while taking into account other relevant public health risks (Suwanrangsi *et al.*, 1999, Shankar and Mohan, 2002). Drugs applied to fish tend to remain in their tissues for longer times as the metabolism is primarily dependent on water temperature. In addition to water temperature, the length of time for elimination of drug and of drug metabolites depends on their factors, such as drug properties, route of application, fish species and its physiological condition, salinity of water. These variable factors make it difficult to set withdrawal periods. At the time of harvesting, veterinary drug residues in fish must not be the maximum possible levels set up by the official agency having jurisdiction (Suwanranghi *et al.*, 1999).

Quality Control and Standards

Quality and safety standards are being explored in various places in order to ensure that capture fisheries and aquaculture operations don't harm the environment (Kourous, 2004). Under eco-labelling system, fish farmed or captured in accordance with certain environmental guidelines are sold with a special label. The responsibility for ensuring the safety of fishery products is shared by every one involved. Science-based safety monitoring and eco-labelling systems are the options for the food safety (Kourous, 2004).

All the quality standards are confined to only processing plants and as such no quality standards exist at shrimp farming sites. To ensure that HACCP is functioning effectively, the pre-requisite program includes Good Manufacturing Practices (GMP). ISO, the International Organization for

Standardization, is a worldwide federation of national standards bodies, comprising of 127 members, representing country including India. ISO develops voluntary technical standards that are required by the market. The ISO 9000 standards are a series of five documents viz. ISO 9000, ISO 9001, ISO 9002, ISO 9003 and ISO 9004. Out of these, ISO 9001, 9002 and 9003 are models for quality assurance is an organization (Iyer and Gopakumar, 2002).

Recent international agreement managed by the World Trade Organization (WTO) has put even further emphasis on the importance of Codex Standards. The objective of Codex is to develop standards for food, protecting the health of the consumers and ensuring fair practices in the food trade. Under WTO, health and safety requirements must be justifiable on grounds of protecting public health and must be based on a sound, scientific assessment. When available, standards from Codex for food safety issues, International Office of Epizootics (OIE) for issues of animal health and International Plant Protection Convention (IPPC) for plant health should be used as references (WHO, 2002).

As the food industry continued to improve quality and production to improve profitability, new management tools were incorporated into companies (Suwanrangi *et al.*, 1999). In 1970, a management tools called quality circles were developed. In the 1980's a management tools called total Quality Management (TQM) became popular. In late 1980's, a management tool was developed to control the flow of goods between nations, the ISO 9000 series standards. The two main roles of ISO 9000 series are to provide guidance for suppliers of all types of products who want to implement effective quality systems in their organizations or to improve their existing quality systems and provide generic requirements against which a customer can evaluate the adequacy of supplier's quality system. During the same period in 1980, HACCP became popular in food industry and in 1990's, it became the national and international quality requirements for trade. Total Quality Management (TQM) is a process of change and improvement in everything the company does. ISO 9000 series requirement are clearly define including

documentation of all work processes that affect quality, but how the requirements are to be met is left to the organization (Suwanrangsi *et al.*, 1999).

Aquaculture product expansion has placed increased requirements on quality and food safety by consumers and regulators. HACCP was adopted by developed countries during the end of 1980s and governments started to shift their regulations to HACCP based system. Some countries have developed comprehensive HACCP plans for selected aquaculture products and in other countries individual aquaculture producers have undertaken voluntary certification (ISO 9000) for control as well as marketing purposes (Josupeit *et al.*, 2001).

2.8 Aquaculture Certification Programs

With the establishment of WTO, food safety has become more prominent and consumers throughout the world are more concerned about antibiotic residues and food borne contamination. There are now several initiatives for certification of shrimp aquaculture and other aquaculture species, and several standards being promoted for shrimp aquaculture.

Alter-Trade Japan (ATJ), a Japanese company developed a certified organic shrimp product, using natural and organic standards (Anon., 2004a). The shrimps were labeled by ATJ as 'eco-shrimps' based on these standards have been exported to Sweden. Aquaculture Certification Council (ACC) was established to certify social, environmental and food safety standards at aquaculture facilities throughout the world. Environmental Justice Foundation (EJF) has prepared a draft protocol for sustainable shrimp aquaculture. Naturland Standards for Organic Aquaculture (NSOA) has developed standards on several aquaculture commodities. Shrimp Seed of Quality (SSOQ), program was established in Bangladesh to certify farmed shrimp, based on code of conduct. Soil Association (SA) has prepared general standards for aquaculture. Thai Quality Shrimp (TQS) of Thailand had prepared codes of conduct for shrimp aquaculture, with the intention of

marketing shrimp produced under the code of conduct with a 'Thai Quality Shrimp' label (Anon., 2003d).

Bio Grass, New Zealand has developed standards for organic fish farming including fish, shell fish, crustaceans and the processing of these products. KRAO (Sweden) and Debio (Norway) have developed general standards for organic aquaculture products in Sweden and Norway.

Bio Suiss, an umbrella organization for organic agriculture in Switzerland is certifying farmed fish such as carp, Cher and Perch and apply only freshwater farming and designed for the Swiss market. Funcacion Chile of chile prepared an environmental code of practice that is intended to serve as the basis for the certification system for Chilean salmon farming. International Federation of Organic Agriculture Movements (IFOAM), a global umbrella body for organic food and farming, has drawn up basic standards for organic aquaculture (Anon., 2004a).

Hazard Analysis Critical Control Point (HACCP)

HACCP stands for Hazard Analysis Critical Control Point. It is a system of food safety management that has been adopted by the USA, EU, Australia and most other countries as the best system to control food safety. HACCP is a set of virtually mandatory requirements in order to export food products. It protects home populations from risky imported food staffs and should be used in all production of food to ensure to safety. Implementation of HACCP has become a requirement rather than a luxury for developing nations and is a legal requirement in most countries like the UK. The International Center for HACCP Innovation (ICHI) in Sanford is the largest HACCP research center in the UK (and probably in Europe) (USFDA, 2001; Anon., 2001 and Tomkin, 1996).

The Pillsbury co-pioneered the application of the HACCP concept to food production during its efforts to supply food for the U.S. space program in the early 1960s. Pillsbury decided that their existing quality control techniques did not provide adequate assurance against contamination during food production. The company found the end-product necessary to provide such

assurance would be so extensive that little food would be available for space flights. To ensure safety, Pillsbury developed a preventive system that kept hazards from occurring during production. Since then, Pillsbury system has been recognized worldwide as the state of the art measure for food safety control. It is not a zero risk system, but it is designed to minimize the risk of food-safety hazards. The FDA first required HACCP controls for food processing in 1973 for canned foods to protect against *Clostridium botulinum*, which cause botulism (Sea Grant, 2001).

HACCP has been endorsed worldwide by organizations such as Codex Alimentarius (a commission of the United Nations) and the European Union and by several countries including Canada, Australia, New Zealand and Japan. HACCP is a preventive system for ensuring food safety, but it is not a stand-alone system. HACCP to be built upon current food safety program such as Good Manufacturing Practices (GMPs) to make it work. Unlike other traditional inspection methods for food safety control, the HACCP approach allows regulators to look at what happens in the plant through time by examining the firm's monitoring and corrective action records (Anon., 2001).

On December 18, 1995, the Food and Drug Administration (FDA) published a final rule 21 CFR 123 "Procedures for the safe and sanitary processing and importing of fish and fishery products" that requires processors of fish and fishery products to develop and implement HACCP system for their operations. The regulations became effective December 18, 1997 (USFDA, 1999).

The HACCP concept proposed by the USFDA has been taken on a standard process control system for assuring food safety by international bodies. Canada, USA, Iceland, European Union and many other fish producing countries have taken to accept as a food safety standard. It has been identified as the global unified quality assurance system for producing safe and better quality fish products at a global level. The FAO's CAC has formulated guidelines for implementation of HACCP system in the food industry. The HACCP concept offers good possibilities to secure the safe production of food. It aims to identify problems before they occur and

establishes measures for their control at all stages in production that are critical in ensuring the safety of food (Lakshmanan, 2002).

HACCP is a worldwide recognized systematic and preventive approach that addresses physical, chemical and biological hazards through anticipation and prevention rather than end product inspection and testing (Iyer, 2003). There is a global shift from food quality to food safety and HACCP is the vital tool to address the food safety issues. The HACCP system works on seven principles (CAC, 1991) and there are 12 steps in the implementation of HACCP. Hazard analysis requires technical expertise and scientific background and it is essential to identify which hazards are of such a nature and their elimination or reduction to an acceptable level is essential for production of a safe food. Critical Control Point (CCP) is the part where hazards need to be prevented, eliminated or reduced to acceptable levels (Antony, 2003).

Hazard analysis is the process of identifying the significant biological, chemical and physical hazards in a process of product. The hazard analysis is critical to develop a complete HACCP plan because hazards, which are overlooked, may not be controlled (Tompkin, 1996). Potential hazards can be identified by using epidemiological records, technical information and hazards. The hazards in seafood can be classified as physical, chemical and biological hazards. Chemical hazards constitute agricultural chemicals that include pesticides, herbicides, animal drugs, fertilizers, etc (Kumar, 2002; CAC, 1991).

Based on the results of a FAO survey regarding the compliance of HACCP procedures, the status of countries has been categorized. Among the developing countries Uruguay, Brazil, Chile, Ecuador, Thailand, India, Malaysia, Philippines, Indonesia, Argentina, Peru, Cuba, Morocco, Sri Lanka, Vietnam and Bangladesh have introduced HACCP procedures. In the second group of countries, the private sector was voluntarily trying to introduce HACCP for fish and seafood export that includes Madagascar, Venezuela, Honduras, Tunisia, Myanmar and Portugal. The third group consists of countries including Russia and China where governments were agreed to

follow HACCP requirements but were still defining the process (Fabres, 2003).

HACCP enables aquaculturists and processors to exercise control over health hazards. It is essentially a technique based upon anticipation and prevention of food safety hazards and it may be applied through out the food chain from producer through final consumer, leading to enhanced food safety and better use of resources (Tookwing and Keerativiriyaporn, 2004).

HACCP is a preventive system of food control that requires a hazard analysis be conducted on the product and process and that critical limits (CL's) are set at each Critical Control Point (CCP) process. In 1991 and 1994, the EU adopted regulations concerning health condition for production and marketing of fishery products and again were roughly based on HACCP principles. Canada was the first country to establish a mandatory food inspection program for fish and fishery products based on HACCP principles (FAO, 2000).

HACCP is a structured, systematic approach for the control of food safety throughout the community system, from the plough to plate. It requires good understanding of the relationship between 'cause' and 'effect' in order to be more pro-active and it is a key element in Total Quality Management (TQM). HACCP builds on foundations of well established Quality Management such as Good Manufacturing Practice (GMP), Good Hygienic Practice (GHP), Good Agricultural Practice (GAP), and Good Storage Practice (GSP) (FAO, 2001b).

During the World Health Organization Study Group on the control of food borne trematode infections held in Philippines in 1993, an attempt was made to design a new strategy of and control of food borne trematodes in cultured fish based on HACCP plans for the production and marketing of fish free from food borne trematode infection (Santos, 1995).

2.7.1 Critical Control Points

Use of Antibiotics is identified one of the critical control point to address the issue of food safety. Required records for every application of antibiotics, drugs and other chemicals shall include the date, compound used, reasons for use, dose and harvest date for treated ponds. Statements from feed and post-larvae suppliers that declare no prohibited antibiotics, drugs or other chemicals were applied to feed or larvae are required. It also suggested that the best ways of controlling diseases in shrimp aquaculture are to avoid stocking diseased post-larvae, reduce water exchange for less exposure to disease organisms in intake water and maintain good bottom soil and water quality to avoid environmental stress to shrimp (ACC, 2004). As aquaculture feeds often make up 50% and more of the production cost, it is clear that research in this field would remain a priority (Sorgelos, 2001). The contribution of feeds to the production costs varies following the species and the system employed, but can be as high as 50% of the total costs (Kongkeo, 2001).

Raghavan (2003) reported that none of the commercial feeds collected from the South Indian shrimp farms have contaminated with any human pathogenic bacteria. The results showed that the total heterotrophic bacteria in the commercial feed samples were in the range of 10^3 to 10^5 cfu/g⁻¹ indicating that the bacterial counts were well within permissible limits. For craw fish production in the USA, 6 steps were identified with pond site selection, pond management and transportation as Critical Control Points (CCPs). For north-west aquaculture fin fish production 6 steps were identified, 3 dealing with site selection/water supply, feed supply and spawning throughout grow out being CCPs. For molluscan shellfish production 10 steps were identified with 8 dealing with approval of site selection, site certification, maintenance, relaying of contaminated products, harvesting, bulk transport, wet and dry storage and packing being CCPs (Garett *et al.*, 2000).

Horowitz and Horowitz (2000) reported that the studies have shown that feed utilization by shrimp is quite poor. Only 17% of the nutrients in feeds of intensive shrimp aquaculture and up in shrimp tissue. About 15% of the

feed is not consumed by the shrimp, and 20% is egested. The largest portion, about 48%, is lost due to molting, maintenance energy, and release by excretion. Thus, the majority of energy in the feed is lost. The reasons for disease out breaks include poor quality of shrimp seeds, failure to adopt better management practices in hatcheries and farms, back of co-operation among farmers is collectively managing the farming activities in farming clusters and careless handling of disease out break. MPEDA/NACA project was started in 2001 in Andhra Pradesh with objectives to find risk factors for shrimp damage. In, 2003 Best Management Practices (BMP's) were implemented in 108 ponds of 58 farmers in one cluster by forming an aqua club. This resulted improvement in yield, and high quality i.e. without any banned chemical residues. Due to implementation of BMPs disease prevalence reduced from 82% to 36% in 2004 (Padiyar, 2005).

The quality of feed was clearly a critical control point since feed influences overall fish performance (daily growth and feed conversion), fish health, the generation of fecal wastes and waste feed, and the total amount of phosphorus ultimately released into the environment. Feed is also a significant cost accounting for 50-60% of production costs (MacMillan *et al.*, 2003).

Shrimp aquaculture has the following critical control points viz. pond site selection, water supply quality, pond management techniques and transportation, especially as it relates to the live transport of aquaculture shrimp species (Garatt *et al.*, 1997). It is essential to consider the unique conditions that exist within a specific fish farm when developing a HACCP plan for it (WHO, 1999).

2.7.2 Application of HACCP in Aquaculture

Aquaculture producers and their feed suppliers need to monitor the quality of their ingredients and feeds constantly to assure they are buying the quality of products they are being sold. A study on application of HACCP in shrimps from pond to plate, by the National Institute of Nutrition (NIN) of Hyderabad found that the samples were contaminated with pathogenic micro-

organisms immediately after harvest and also at the market place. The recipes prepared from prawn were free from pathogenic organisms while pond water samples were contaminated with micro-organisms including *Vibrio* sp., feed and ice samples used for preserving were free from pathogenic organisms. Results of the physical contaminants revealed that all the prawn samples collected at the pond site after harvesting and after reaching the markets of Hyderabad city contained 2-4 pieces of scales and none of the recipes prepared from prawns contained the physical contaminants. The study emphasized that the HACCP process should not be confined only to processing unit but need to be extended to the entire food chain from pond to plate (NIN, 2004).

The HACCP system is recommended as a way of reducing hazards stemming from the processing of fish and fishery products. Implementation of the HACCP system in fish processing is mandatory, and all exporting countries have to comply with this requirement for international trade. Since the middle of 1990, some developed countries have introduced the system to control hazards from the use of antibiotics at the pond level (Vaslet, 1997).

The introduction of HACCP to control food hazards in aquaculture, including those stemming from the irresponsible use of antibiotics has been widely recommended (Reilly et al., 1997) and has been discussed by an FAO/Network of Aquaculture Centres in Asia-Pacific (NACA)/WHO Study Group on food safety (WHO, 1999).

HACCP is currently not mandatory by most primary animal production regulations that include aquaculture. In many countries, even when the liability may be shared or (depending on regulations) when it remains on the production side, the actual obligation to control the use of antibiotics and their residues rests with the processing industry, as HACCP is mandated within the processing sector. This creates difficulties in implementing control measures on antibiotic use in aquaculture (FAO, 2002).

All the elements for identifying the critical control points (CCPs) and critical limits of regulatory requirements exist for approved antibiotics and

veterinary drugs, specific fish or shellfish species, diagnosis (purpose of use), dose, duration of treatment and withdrawal period. It has been suggested that the CCP would be at the feeding stage, since this is when antibiotics are usually introduced into the production process. The analysis of residues of the antibiotics used, and the checking of compliance with regulations, would form part of the verification procedures. In addition, as USFDA has suggested, the monitoring of residues in flesh may be not enough, and the development of resistance in pond micro-organisms (and/or the target micro-organism) should also to be monitored, an additional CCP. Application of HACCP – based management practices within production system in central for reducing possible risks. Appropriate guidelines and technical standards should be developed in consultation with all stakeholders. There is also a need to reassure consumers about the safe use of approved antibiotics and measures to constrain the use of banned substances (FAO, 2002).

During a study conducted at Cochin and Mumbai on the quality of fish sold in retail markets, *E. coli* was detected in 98.7% and 96.85% of the fresh and frozen fish samples respectively; these results were indicative of the need for introduction of good handling practices and quality standards for fish and fishery products for international markets and emphasized the importance of the implementation in order to safeguard the health of the consumers (Joseph *et al.*, 1983, 1988; Kalaimani *et al.*, 1988; Valsan *et al.*, 1985; Iyer and Srivastava, 1988; Latha and Lakshmanaperumalsamy, 1997; Nambiar and Iyer *et al.*, 1990, Nambiar and Surendran, 2003).

The HACCP concept applies from production to consumption. The aquaculture products might represent some unique risks from antibiotic residues, herbicides, etc. An expansion of these hazards would include pesticides, microbial pathogens and chemical contaminants and unapproved food additives, as also veterinary residues including hormone, growth regulators, and antibiotics (Tookwinas and Keerativiriyaporn, 2004; Tookwinas and Suwanrangsi, 1996). At the production step, the three possible hazards were related to misuse of registered or non-registered chemicals or drugs. Non-conforming antibiotic residues in flesh were due to

inadequate withdrawal times, and/or pathogen contaminates of the fish (Garott *et al.*, 2000). The preventive measures necessary to control the hazards at the production step were identified as (1) the use of approved drugs and chemicals at proper concentrations (2) executing drug withdrawal procedures from feeds in accordance with antibiotic labeled instructions and (3) prohibiting use of feed or water contaminated with hormone and/or animal wastes (FAO, 2002).

Preventing hazards to peoples' health from cultured products can be more effective by the application of HACCP at the farm, in other words, before the raw material even goes into the processing plant. The Department of Fisheries, Thailand had developed preventive approaches to assure control over raw material, the manufacturing process, the production environment, and personnel. It is based on the identification of potential hazards, application of control measures at critical control points (CCP) and monitoring and verifying of CCPs there by enabling the assurance of food safety during culture and processing (Tookwinas and Keerativiriyaporn, 2004).

Hartog (2004) reported that GMP and the quality assurance system used by the Dutch animal feed sector were considered as one of the best and most rigorous systems of its type in the world. Transforming GMP regulations from a reactive to a pro-active system by introducing HACCP in the quality system (GMP+), whereby risk assessment is carried out first and preventive measures are established base on assessment. Implementation of HACCP in the animal feed sector is virtually complete. Quality assurance in the raw material chain is also an advanced phase of implementation Hartog (2004).

Water supply, fish ponds, harvest, village market and retailer, processing plant, home and restaurant were identified as critical control points (Santos, 1995). Applications of HACCP principles were recommended to address the food safety issues associated with products from aquaculture (WHO, 1999). Hazards can enter the food chain on the farm and can continue to be introduced or exacerbated at any point in the food reaches the consumer. The holistic approach to the control of food related risks involves

consideration of every step in the chain, from raw material to food consumption (WHO, 2002).

HACCP was widely used in food processing industry especially fish and fishery products. There are some initiative studies for apply HACCP to fish production sectors, in order to ensure the safety of raw material. HACCP will be most effective it applied across a continuous, popularized by the expression 'farm to fork". This approach implies that HACCP must start at the production (pond) level and continue though to consumer education. Once a fish farm has established a pre-requisite program, the principles of HACCP can then be applied at each individual production process. The HACCP is meant to serve only as a model and an establishment, must prepare a plan for the specific conditions prevailing in production unit, and for a particular product. While application of HACCP is well advanced in the fish processing sector, the application of HACCP in aquaculture and some other part of food chain that include handling and transportation, are in infancy (Suwanrangi *et al.*, 1999). The aquaculture sector is not unique in this respect and there are certain stages of the food chain that are ideally suited to the application of HACCP principles. In applying HACCP it is necessary to carefully examine the nature and extent of any hazards associated with methods of production and products from aquaculture (Reilly and Kaferstein, 1997).

The project work conducted by Gunderson and Kinnun (2001) had yielded positive results by implementing HACCP principles to prevent the spread of aquatic nuisance species in aquaculture and bait fish operation.

Jahncke *et al.* (2000b, 2001a, 2002) reported that HACCP principles were applied as a risk management tool to control viral pathogens at shrimp aquaculture production sites and a shrimp processing facility. At each operational step and CCP, hazards were identified and assessed, control measures and critical limits were established, and monitoring procedures and record keeping requirements were identified. Outsourced live shrimp shipment of live and finished product, feed receipt, and incoming water were identified as CCP, as well as effluent water at the pond grow out step.

HACCP principles provide a logical step-wise approach to identify and control animal and environmental hazards associated with aquaculture production and processing facilities. This management approach emphasizes process control and concentration on the points in the operation that are critical to animal and environmental safety (Jahncke *et al.*, 2001a, b; 2002).

The statement of the Government of Thailand, prepared for the Aqua Markets – 2003, held in Philippines, informed that it started working on the implementation of HACCP principles at the shrimp farm sites as the first critical control point is the raw material. A recommendation on this aspect is for a study in Thailand and or Asia to estimate the cost and benefits of producing the raw material that meet quality and safety criteria established by processors and importing countries (Anon., 2003d).

The potential critical control points in catfish farming are water supply, feed supply and production (Smith, 2002). HACCP principles have been applied to catfish producers in USA. The incorporation of HACCP system into the fish health program of an aquaculture facility is one way to help guarantee a safe and wholesome product for both the producer and consumer of cultured finfish. The implementation of HACCP would also help insure consistent marketable products. This system, like that used in seafood safety regulations, is based on the analysis of potential health hazards for the fish and human consumer (Smith, 2002).

Individual facility specific Best Management Practice (BMP) plans were developed using a HACCP like approach. These efforts have resulted in a 40% reduction in effluent phosphorus from measured 1990 man loads. Fish production volumes and fish quality have been maintained and costs limited (MacMillan *et al.*, 2003).

HACCP principles can also be applied to feed manufacturing to assure the quality of aquaculture feed manufacturing to assure the quality of aquaculture feed by providing a systems approach in which steps to improve feed quality can be identified and prioritized. Feed manufacturing using this approach can significantly improve the quality of their feeds (Hardy, 1991).

The FAO and WHO recommended that the HACCP concept be applied to freshwater aquaculture program in Asia to control food borne digenetic trematode infections in humans. In a study in Vietnam, experimental activities were conducted in two side-by-side fish ponds. In experimental ponds, fish were cultured in conjunction with HACCP principles and control pond was cultured according to conventional local aquaculture practices. Water supply, fish fry, fish feed and pond conditions in the experimental pond were identified as critical control points. The basic HACCP principles were applied. The results showed that pond which applied HACCP principles were completely free of trematode infection whereas 40% control pond infected with *Clonorchis sinensis*. The results indicate the application of HACCP – based principles to silver carp culture in North Vietnam was an effective way to prevent and control *C. sinensis*. Similarly the application of this principle to freshwater aquaculture ponds in Thailand and Laos to control *Opisthorchis viverrini* infections had also been successful. Application of HACCP principles at aquaculture site locations has the potential to control transmission of exotic human and animal pathogens (Garett *et al.*, 1997).

Application of HACCP approach in other food sectors

Rathi (2002) have applied HACCP principles at mushroom producing unit located in the outskirts of Hyderabad, Andhra Pradesh, India. The HACCP concept has been successfully applied in the control of quality as well as safety in low acid canned foods in the USA, and many food companies in Europe and the USA have adopted the approach. Increasingly, regulatory bodies have recognized usefulness of this tool and its 'principles' have been incorporated into legislative requirements by both the EU in the General Hygiene regulations for managing food safety (93/43/EEC), and the United States Federal Department of Agriculture (CPR-123). The National Advisory Committee on Microbiological Criteria for Foods (NACMCF) provided guidelines on HACCP including generic plans and decision tree in 1992, and the Codex Alimentarius Commission adopted the HACCP system at its twentieth session in 1993. HACCP system can be incorporated into other quality assurance systems such as the ISO 9000 series (FAO, 2001a,b).

HACCP principles have been applied to control mycotoxins (FAO, 2001c). Codex Alimentarius approved HACCP system in 1993 for adoption by every food producing sector. The study conducted in Thailand, to apply HACCP and risk based approach for food safety strategy to Somtam, a street vended food in Thailand revealed positive results (Anon., 2000a&b).



Materials and Methods



3. MATERIAL AND METHODS

3.1 Field Survey at Coastal Shrimp Farms of India

In order to develop a HACCP frame work feasible to Indian shrimp farming, in compliance with 'Codex' standards (1991), a field survey had been conducted to have hands-on information on existing shrimp aquaculture practices, for identification of Critical Control Points (CCP's), and to analyze the factors responsible for the hazardous substances like antibiotic residues in harvested shrimp, so that the data could provide support development of a suitable HACCP framework at the production level.

3.1.1 Location of the Study

The study was conducted during the year 2003-04 at 120 shrimp farms located in the coastal districts of Andhra Pradesh and Tamil Nadu on the east coast and Gujarat, Maharashtra, Karnataka and Kerala on the west coast of India. The survey in Andhra Pradesh was carried out in the districts of Nellore, East Godavari, West Godavari, Vishakhapatnam, and Prakasham, while that in Tamilnadu was carried out in the districts of Pattukottai, Kancheepuram and Nagapattinam. Thane district in Maharashtra, Surat district in Gujarat, Udipi district in Karnataka and Ernakulam district in Kerala were selected on the west coast based on the extent of area shrimp farming, level of production and reported problems.

3.1.2 Population and Sample

A total of 120 respondents were selected whose details are given in Table 1.

Table 1. Details of States and Respondents involved in Field Survey

Sl. No.	Name of the State	Name of the District	Total No. of Respondents identified	No. of Respondents selected
East-coast				
1.	Andhra Pradesh	Vishakhapattinam, East Godavari, West Godavari, Nellore, and Prakasham	200	45
2.	Tamil Nadu	Pattukottai, Kancheepuram and Nagapattinam	150	20
West-coast				
3	Maharashtra	Thane	65	12
4.	Gujarat	Surat	80	15
5.	Karnataka	Udipi	80	13
6.	Kerala	Ernakulam	50	15

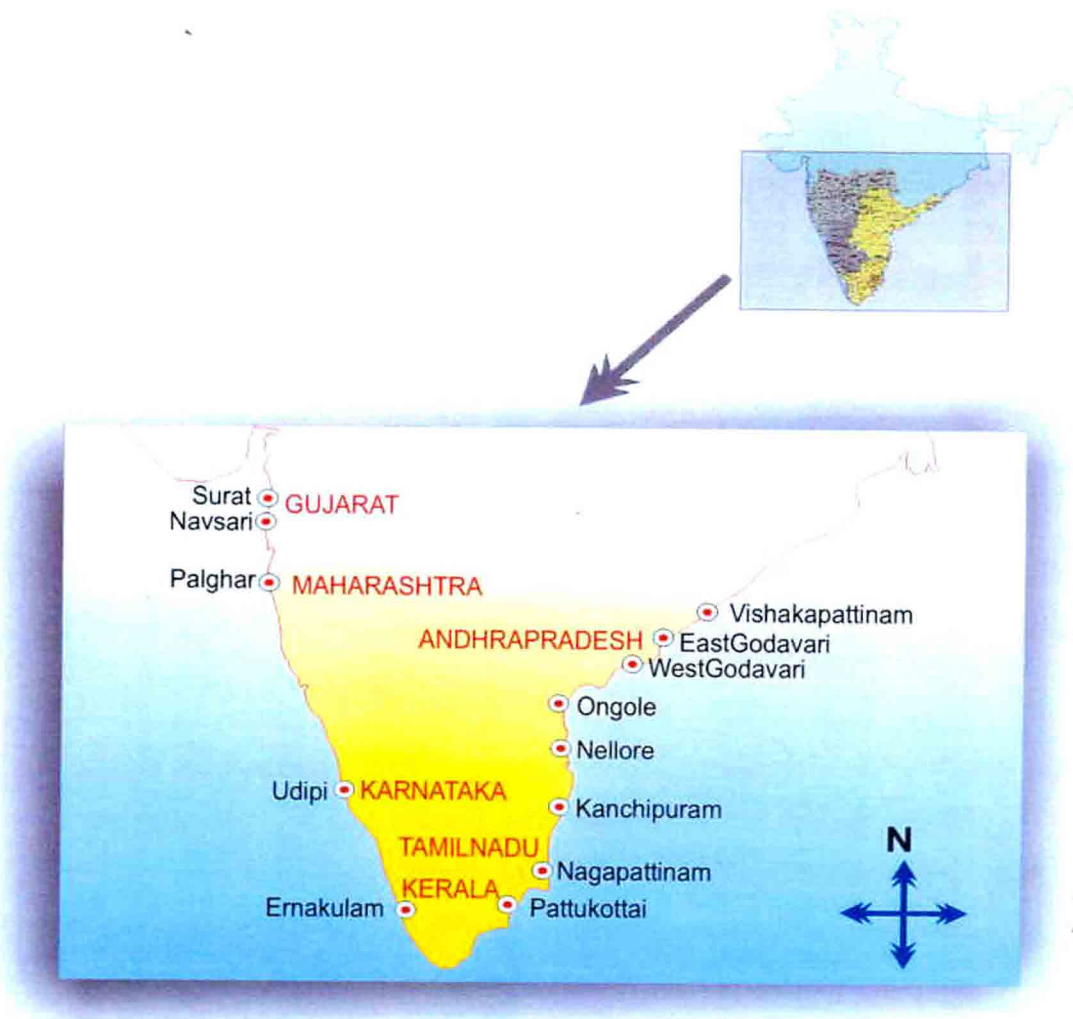


Fig. 1 Map showing the study area

3.1.3 Research Design and Method

Ex-post-facto research design was used, which according to Robinson (1976) is a systematic empirical inquiry in which the independent variables have not been directly manipulated because they have already occurred or because they are not inherently manipulable.

The data was collected with the aid of a well-structured interview schedule and was conducted through personal interview, as it was considered appropriate for acquiring necessary information first-hand from the farmers.

3.1.4 Operationalisation and Measurements of variables

The variables used for the present study was broadly categorized into Profile characteristics of shrimp farmers and shrimp farms, characteristics of intensity of shrimp farming, shrimp farm management practices, sources of seed and feed, chemicals used in shrimp farming, characteristics of marketing, technical awareness related to shrimp farming, diseases in shrimp farming, problems in shrimp farming, reasons for crop loss and farmers suggestions for better results in shrimp farming.

Out of the total 46 variables prepared, 31 variables were finalized and included in the present study based on review of literature, discussion with faculty members and subject experts of CIFE, Mumbai; NAARM, Hyderabad; and ANGRAU, Hyderabad. A set of statements in the form of questions, some open ended and others with relevant multiple answers were developed systematically and pre tested.

3.1.4.1 Profile of Characteristics of Shrimp Farmers and Farms

The profile of characteristics of different shrimp farmers and farms were categorized and scoring pattern adopted as follows:

3.1.4.1.1 Shrimp Farmer Education: It refers to the number of years of formal education acquired by the respondent. The education level of response categories and scoring pattern adopted in the present study are as follows:

<i>Education</i>	<i>Score</i>
Illiterate	0
Primary school	1
Matric / Higher Secondary school	2
Graduate	3
Professional	4

3.1.4.1.2 Occupation: It refers to the occupation, either main or secondary, of the respondent. The scoring pattern adopted is as follows:

<i>Occupation</i>	<i>Score</i>
Aquaculture + Agriculture	1
Aquaculture only	2
Aquaculture + Aqua-business/others	3

3.1.4.1.3 Shrimp Farming Experience: It refers to the number of years since the respondent had been involved in shrimp farming activities at the time of enquiry. The scoring pattern adopted is as follows:

<i>Experience</i>	<i>Score</i>
No experience	0
One year	1
Two years	2
Three years	3
More than three years	4

3.1.4.1.4 Farm Type : It refers to the pattern of land ownership of the respondent. The scoring pattern adopted is as follows:

<i>Farm Type</i>	<i>Score</i>
Leased	1
Own	2

3.1.4.1.5 Soil Type: It refers to the type of soil present in the shrimp farms. The scoring pattern adopted is as follows:

<i>Soil Type</i>	<i>Score</i>
Sandy loamy	1
Sandy clay	2
Clay loam	3

3.1.4.1.6 Farm Area: It refers to the area in hectares, used for shrimp farming. The scoring pattern adopted is as follows:

<i>Farm area (ha)</i>	<i>Score</i>
Less than 2	1
Between 2 and 5	2
More than 5	3

3.1.4.2 Characteristics of Intensity of Shrimp Farming

The characteristics of intensity of shrimp farming were categorized and scoring pattern adopted as follows

3.1.4.2.1 Stocking Density: It refers to the number of animals stocked per square meter. The scoring pattern adopted is based on the assumption that higher the stocking density, higher the probability of occurrence of problems are higher.

<i>Stocking Density (no/m²)</i>	<i>Score</i>
Less than 15	1

13-15	2
8-12	3
5-7	4

3.1.4.2.2 Months of Stocking: It refers to the usual months of stocking the post-larvae of shrimp. Scoring pattern is adopted based on the best months preferred for optimum yield.

<i>Month of Stocking</i>	<i>Score</i>
January or February or March	2
June or July or August or September	1

3.1.4.2.3 Culture Period: It is the duration of grow-out period before harvesting. The scores have been allotted based on the marketable size achieved, cost of production and profit.

<i>Culture Period (days)</i>	<i>Score</i>
Below 120	1
120 – 150	2

3.1.4.2.4 Number of Crops per Year: It refers to the number of crops taken in a year. The scoring pattern adopted is as follows:

<i>No. of Crops/Year</i>	<i>Score</i>
One	1
Two	2
Three	3

3.1.4.2.5 Crop Rotation: It refers to the farming of organisms other than shrimp *i.e.* fish or prawn in the same pond. The scoring pattern adopted is as follows:

<i>Crop Rotation</i>	<i>Score</i>
Yes	1
No	2

3.1.4.2.6 Aerators: It refers to the use of mechanical devices to increase the dissolved oxygen levels in the pond. The scoring pattern adopted is as follows:

<i>Aerators</i>	<i>Score</i>
No	0
Yes	1

3.1.4.2.7 Water Exchange: It refers to the percentage of pond water exchanged during shrimp culture and the number of times such an exchange was carried out per month. The scoring pattern adopted is as follows:

<i>Water Exchange</i>	<i>Score</i>
Percentage of Exchange:	
<10 and >15	1
10-15%	2
Number of Times (Exchange once in) :	
No exchange	0
30 days	1
15 days	2
7 days	3

3.1.4.2.8 Water Depth: It refers to the depth of the water in the shrimp farm. The scoring pattern adopted is as follows:

<i>Water Depth (m)</i>	<i>Score</i>
1.0 -1.5	2
Less than 1	1

3.1.4.2.9 Water Source: It refers to the water drawn for the culture of shrimp. The scoring pattern adopted is as follows:

<i>Water source</i>	<i>Score</i>
Ground water	1
Creek and surface water	2

3.1.4.3 Shrimp Farm Management Practices

It refers to the adoption of prescribed optimum culture conditions that were followed in shrimp farms. The optimum culture standards prescribed by MPEDA and other Government organizations were taken into account while allotting scores to the different parameters

Culture Conditions

S.No	Parameter	Values	Score
1	pH	7.5 - 8.5(optimum)	2
		6.0 - 7.5 and 8.5-11.0	1
		<6.0 and >11.0	0
2	Salinity (ppt)	15 to 25(optimum)	2
		5 - 15 or 25 - 30	1
3	Transparency (cm)	30-40 (optimum)	
		≤30 and ≥40	
4	Dissolved Oxygen (ppm)	≥5 and above (optimum)	2
		3 - 4	1
		≤3	0
5	Plankton	Medium (optimum)	3
		Rich	2
		Poor	1
6	Water Colour	Green brown or yellow brown	2
		Other Colour	1
7	Frequency of testing parameters	7 days (optimum)	3
		15 days	2
		30 days	1
		No testing	0

3.1.4.4 Characteristics of Seed and Feed:

The characteristics of the sources of seed and feed used in shrimp farming were categorized and scoring pattern adopted as follows:

3.1.4.4.1 Source of Seed: It refers to the source of post larva used for stocking in the grow-out operations of shrimp farming. The scoring pattern adopted is as follows:

<i>Source of seed</i>	<i>Score</i>
Hatchery	2
Wild	1

3.1.4.4.2 Age of Post-Larvae: It refers to the age in days after completing larval phase. The scoring pattern adopted is as follows:

<i>Age of the PL (in days)</i>	<i>Score</i>
PL more than 20 and less than 15	1
PL 15-20	2

3.1.4.4.3 Seed Testing : This refers to the testing of post-larvae, which is used for stocking in grow-out ponds. The scoring pattern is followed based on the type of tests carried out before stocking.

<i>Seed testing</i>	<i>Score</i>
No test	0
Stress test	1
PCR test	2
All tests	3

3.1.4.4.4 Source of Feed: It refers to the source of supplementary feed used for the shrimp culture and feed conversion ratio of the feed used. The scoring pattern adopted is as follows:

<i>Source of Feed</i>	<i>Score</i>
Local	1

Local and branded	2
Only branded	3

3.1.4.4.5 Feed Conversion Ratio (FCR): It refers to the ration of supplementary feed consumed (kg) for yielding 1kg of body weight of shrimps. The scoring pattern adopted is as follows :

Feed Conversion Ratio (FCR)

> 1.8	1
1.6 - 1.8	2
<1.5	3

3.1.4.5 Chemicals used in Shrimp Farming: It refers to the chemicals and drugs used by the respondent during shrimp culture practices. The scoring pattern is 'Yes', 'No' and 'No response'. The scoring for 'No response' was included to study the extent to which respondents are forthcoming to answer questions related to the use of chemicals, since it had been reported that though the farmers use antibiotics, they denied the fact when they were confronted.

<i>Chemicals</i>	<i>Score</i>
(a) <u>Antibiotics</u>	
No response	0
Yes	1
No	2
(b) <u>Sanitizers</u>	
No response	0
No	1
Yes	2

(c)	<u>Molt inducers</u>	
	No response	0
	Yes	1
	No	2
(d)	<u>Probiotics</u>	
	No response	0
	No	1
	Yes	2

3.1.4.6 Characteristics of Marketing: It refers to the selling of the harvested shrimp. The scoring pattern adopted is as follows:

	<i>Marketing</i>	<i>Score</i>
(A)	Agent	1
	Direct to Company	2
(B)	Head less	1
	Head on	2

3.1.4.23 Technical Awareness about Recent Developments

This refers to the extent of knowledge and awareness of the respondent on latest developments with respect to sustainable aquaculture practices and production of quality raw material. To test the awareness level, training need, willingness to adopt new technology, questions with appropriate scoring pattern were developed which are as follows:

	<i>Items</i>		<i>Score</i>
(a)	Are you aware of banned antibiotics?	Yes	1
		No	0

(b)	Are you aware of MPEDA guidelines on usage of antibiotics in shrimp farming ?	Yes	1
		No	0
(c)	Are you aware of MPEDA Code of Conduct of Practice?	Yes	1
		No	0
(d)	Have you undergone any training related to shrimp farming?	Yes	1
		No	0
(e)	Do you need any training related to shrimp farming?	Yes	1
		No	0
(f)	Are you willing to adopt any new technology for improving the quality of shrimp ?	Yes	1
		Yes	1
		No	0

3.1.4.7.1 Source of Technical guidance: It refers to the source of technical guidance to the respondents. The following scoring pattern was used:

<i>Technical guidance</i>	<i>Score</i>
Self	1
Feed company technician	2
Scientists (or) qualified professionals	3

3.1.4.8 Diseases in Shrimp Farming: It refers to the occurrence of diseases experienced by the respondent during the present shrimp farming. The farmers were asked to list out all the. The reported diseases were then categorized and ranked accordingly.

3.1.4.9 Problems in Shrimp Farming

It refers to the major problems experienced by the respondent during shrimp farming. The farmers were asked to list out all the problems associated with shrimp farming. The reported problems were then categorized and ranked accordingly.

3.1.4.10 Reasons for crop Loss:

It refers to the reasons for the loss of crop. The reasons were classified into seven categories and the following scoring pattern was used to identify the most important reasons for crop loss.

	<i>Reasons for crop loss</i>		<i>score</i>
(i)	Seed	Yes -	1
		No -	0
(ii)	Water quality	Yes -	1
		No -	0
(iii)	Feed quality	Yes -	1
		No -	1
(iv)	Climate	Yes -	1
		No -	0
(v)	Poor management	Yes -	1
		No -	0
(vi)	Poor technology	Yes -	1
		No -	0
(vii)	Diseases	Yes -	1
		No -	0

3.1.4.11 Farmers Suggestions for Better Results

It refers to the suggestions provided by the respondents for getting better results in terms of high yield, and quality product. The farmers were asked to provide suggestions as per their own experience in shrimp farming. The suggestions were categorized accordingly and ranked.

3.1.5 STATISTICAL ANALYSIS

The collected data from the respondents were scored, tabulated and analyzed using SPSS.11 version of computer program. The following statistical tools and techniques were used for the analysis of data.

3.1.5.1 Frequency and Percentage

The data were presented in frequency and percentage to understand the nature of the distribution of shrimp farmers and to know the practical problems in shrimp farming, reasons for crop loan and suggestions received from the farmers for yielding better results.

3.1.5.2 Cross Tabulation Table

To test the relationship between two categorical variables of collected data, the cross tabulation data was used.

3.1.5.3 Kendall's tau –b and tau – c

The data was analyzed for symmetrical and directional measures of ordinal association based on the idea accounting for concordance versus discordance.

3.1.5.4 Spearman Rank Order Correlation Coefficient

To measure the association at ordinal level, Spearman's rank order correlation coefficient was used on the collected data. This is a non-parametric version of the Pearson's correlation based on the ranks of data rather than the actual values.

3.2 Estimation of Oxytetracycline Withdrawal Period in *Penaeus monodon*

OTC is an approved antibiotic in India with a Maximum Residue Limit of 0.1ppm in the raw material (GOI, 2002). As there is no antibiotic available to control specific microorganisms, Oxytetracycline (OTC), a broad-spectrum

antibiotic is most commonly used in shrimp farming and hatchery operations to control endemic and systemic Vibrios through medicated feed. Keeping in view of the frequency of its usage and importance in shrimp farming practices, OTC was selected for estimation of withdrawal period for the black tiger prawn *Penaeus monodon* under given conditions. The ambiguity of data on withdrawal period with respect to Indian conditions was the reason for the present study. To fix the critical limits for the identified critical control points, OTC was used for the estimation of withdrawal time, under the given conditions.

3.2.1 Tank Preparation

Four FRP tanks each of 1500 l capacity and measuring 0.75 m in height and 1.8 m in diameter were used for the study. Among the four tanks, three were used for triplicate set of experiments and one as control. 15 days prior to the commencement of experiment, seawater was collected from the nearby rocky beach during high tide time and kept in a separate tank of 2,000 l capacity and allowed for settlement for one week. The upper column of the water was transferred to the four tanks. Small amounts of alum and liquid chlorine were applied for conditioning of the water and proper aeration was provided for all the tanks. The salinity of the water was maintained at 13 ppt, in line with the pond where shrimps were collected.

3.2.2 Shrimp Collection

300±15 numbers of 80 days old shrimps of average body weight of 10g were collected from M/s. Pancham Aqua, Safale, Thane (Dt), Maharashtra. The shrimps were collected from the ponds where antibiotics were not used. The animals were brought under oxygen to the CIFE Aquaculture laboratory, Mumbai. Salinity of pond water at the farm and the transport time were noted. After reaching the experimental station, shrimps were transferred immediately to the experimental tanks at the rate of 50 animals per each tank.

3.2.3 Rearing of Shrimps

Semi-intensive culture practices were followed throughout the experiment. Salinity, pH and Dissolved Oxygen were maintained at optimal levels. Shrimps were acclimatized for 10 days and the medicated feed was started from the 11th day onwards for a period of 7 days and post medication for 18 days. 'The Water Base (TB4)' feed was used for the entire period of study.

3.2.4 Feed Medication

Oxytetracycline Soluble powder (250 mg of OTC active ingredient in each 4 gm veterinary grade of M/S Pfizer Animal health, Navi Mumbai) was used for the present study. 5g of OTC dissolved in 45 ml of distilled water was sprayed on 1 kg of feed to obtain OTC concentration of 5 gm / kg of diet. The experimental diet was prepared on a daily basis. To protect leaching of the OTC, pure cod liver oil was added at the rate of 30 ml per kg feed. Feed was given 4 times daily at the rate of 3% body weight during 0630, 1130, 1730 and 2330Hrs. Medicated feed was given for 18 days and non-medicated feed was given for the remaining 18 days of culture. Control tank received only non-medicated feed throughout the experiment.

3.2.5 Sampling procedure

Prior to second feeding time, 3 animals from each tank were collected randomly every day from the first day of starting of medication till the end of the experiment. Twelve animals were sampled every day and the length and weight were measured and the animals were observed for symptoms of disease if any. Animals were sealed in different polythene pouches, labeled and kept at -20°C until further analysis.

3.2.5 Sample analysis

The samples collected during the medication period (1-7 days) and post-medication period (8-25 days) were analyzed using a High Pressure Liquid Chromatography (HPLC).

3.2.5.1 Sample preparation

The method of Ueno and Aoki (1999) was followed with slight modifications. 1g of muscle tissue pooled from 3 animals from each tank collected each day was homogenized with 5ml cold 30% Methanol containing 0.5% EDTA in 10 ml tube, and centrifuged at 4,000 rpm for 5 minutes. Supernatant was filtered through a syringe filter unit of 0.2m diameter.

3.2.5.2 HPLC analysis

All the aliquot samples of experimental tanks and control were taken in a insulated box to Therapeutic Drug Monitoring Laboratory, Sion, Mumbai for HPLC analysis. The mobile phase used was Methanol-0.2 M Oxalic acid (1:9v/v); Seepak 'C18' cartridge was used as column for extraction using UV detector. The standard solution was 100µg/ml kept in water at -20°C. Aliquots of samples (100 µ l) were injected into the HPLC.

3.3 Hazard Analysis of Shrimp Feeds

Microbiological and physical hazard analyses of shrimp feeds were carried out to assess the level of contamination by microbiological and physical hazards (USFDA, 2001).

3.3.1 Microbiological Hazards

The samples were analyzed according to the Mackie (1989), for the presence of Coliforms, and for Total Plate Count (TPC).

3.3.1.1 Collection of Feed Samples

Samples of feed manufactured by 10 different feed companies were randomly collected aseptically from the presently studies shrimp farms located at Nellore, East Godavari, and West Godavari districts of Andhra Pradesh where shrimp farming is practiced on a large scale. For each brand, samples were collected from 3 different farms; thus a total of 30 samples were collected. Each brand was given one code, thus feed samples A – J. The details

are given in Table 2. All the collected feeds were manufactured 1-2 months prior to sampling and the feeds were using during 90 days of culture.

3.3.1.2 Processing of Samples

1g of each feed sample was taken aseptically to which 9ml of 0.1% peptone water (pre-sterilized diluent) was added.

3.3.1.2.1 Peptone water

0.1% peptone water was used for preparing serial dilutions of samples.

(A) Composition

Peptone	:	0.2 g
Sodium Chloride	:	1.0 g
Distilled water	:	200 ml
pH	:	7.2

(B) Preparation

200 mg of peptone was added with 1g of Sodium Chloride and dissolved in 200 ml distilled water. Adjusted the pH to 7.2 and was dispersed in each tube, plugged and autoclaved at 15 lbs for 15 minutes. All the feed samples were mixed well in the diluent. From the stock, serial dilutions (10^{-1} , 10^{-2} , and 10^{-3}) were prepared in 0.1% peptone water. An aliquot of 100 μ l of all serially diluted samples were plotted out and spread on respective media using surface plating, standard plate count (SPC) method.

Table 2. Details of Feed Samples Collected for Hazard Analysis

S.No	Feed Sample	Details of Feed	
		Manufacturer*	Brand Name*
1	A	Avanti-Pingtai	Profeed
2	B	Higashimaru	Top feed
3	C	Godrej	Kattochi
4	D	The Waterbase	Tiger Bay
5	E	Gold Coin	Gold Forta
6	F	Gold Mohor	Hanaqua
7	G	Lila Global	Global Star
8	H	CP India	Shrimp Feed
9	I	M.N.R aqua Feeds	Maico Gold shrimp feed
10	J	Grobest	Smart prawn feed

*This study is not an endorsement for any commercial product

Pre poured hardened agar plates with dry surfaces were employed. Selective media (agar) for each type of bacteria to be enumerated was used as described in Table 2. The commercial media was obtained from M/s. Hi media, Mumbai and formulation and preparation of agar plates were done as per the manufactures specifications. Samples were spread on total surface of the media evenly using sterile spreaders (bent glass rods). The plates were then incubated at recommended temperature. Colonies were counted after the incubation time was completed. Conditions and time for each media are mentioned in Table 3.

3.3.1.2.2 Bio-chemical tests

(A) Indole test

This test was used to determine the ability of certain bacteria to decompose the amino acid tryptophan to indole, which accumulates in the medium. Indole was then tested by a calorimetric reaction with p-dimethyl amino benzyaldehyde.

Materials

Tryptone – 10 g

Sodium chloride – 5 g

Distilled water – 1000 ml

pH adjusted to 7.4 and sterilized by autoclaving at 121⁰C for 15 minutes

Kovac's reagent

p-dimethyl amino benzyaldehyde – 10g

Hydrochloric acid (conc) – 50ml

Amyl or isoamyl alcohol – 150 ml

Procoduro

The broth was inoculated with the organism taken from an isolated colony and incubated at 37⁰C for 48 hours. 0.5 ml of Kovac's reagent was added to the culture fluid (about 6 ml) and mixed by rotating the plate between the palms slowly. A red colour in the alcohol indicates positive reaction.

Total aerobic plate count

$$\text{TPC per gram} = \frac{\text{Number of colonies} \times \text{dilution}}{\text{Weight of the sample}}$$

Total Coliform count

$$\text{TCC per gram} = \frac{\text{Number of colonies} \times \text{dilution}}{\text{Weight of the sample}}$$

3.3.2 Physical Hazards

The collected samples were observed for the physical hazards specified by the USFDA (2001).The texture of the feed samples were also examined to analyze the binding capacity of the pellets which reflects the procedures adopted in manufacturing.

Table 3. Types of Media and Incubation Time for the Microbiological Hazard Analysis of Shrimp Feeds

S.NO	BACTERIA	MEDIA	INCUBATION	
			TEMPERATURE (° C)	TIME (hrs)
1	Aerobic plate count	Nutrient Agar	37	24
2	Coliforms	Mac Conkey Agar	37	24
3	<i>S.aureus</i>	Mannitol salt agar	37	48

3.4 Development of a HACCP Framework for Preventing Antibiotic Residues from Indian Shrimps

The seven principles, used to establish and maintain a HACCP plan. The HACCP principles have been published by the Codex Alimentarius Commission, (1991) consists of the following principles :

- i. Identification of Hazards;
- ii. Establish Critical Control Points;
- iii. Set Critical Limits;
- iv. Monitor;
- v. Take corrective Action;
- vi. Keep good records; and
- vii. Verify success.

3.4.1 Steps for Developing HACCP Framework

The above seven principles of HACCP form the basis for developing a framework consisting of the following 12 steps, based on Suwanrangse *et al.*(1999).

- i. Assembling HACCP team
- ii. Describing the produce/product
- iii. Identification of end users

- iv. Construction of a flow diagram
- v. On-site verification of the diagram
- vi. Conduct Hazard analysis
- vii. Identification of Critical Control Points (CCP's)
- viii. Establish Critical Limits for each CCP
- ix. Establish Monitoring procedures
- x. Establish Corrective Action procedures
- xi. Establish Verification procedures
- xii. Establish Documentation procedures

Based on the above, suggestions / recommendations were arrived at for developing the said HACCP Framework at the farm level. Further, a model flow Diagram was also developed to fulfill the need as at Step 4 and tested as at Step 5.



Results

4. RESULTS

4.1 Field Survey at Coastal Shrimp Farms of India

4.1.1 Profile of characteristics of Shrimp Farmers and Farms

The results pertaining to profile characteristics are presented in Tables 4(a) and 4(b).

4.1.1.1 Education level of Shrimp Farmers

The results reveal that on a national basis 40% of the shrimp farmers were SSC and Higher Secondary (HSC) passed in overall India and mainly west coast region farmers representing the major share of 50.9% compared to east coast region, which was 32.3% only. Amongst the west coast region the shrimp farmers in the Karnataka state represented more 76.9% farmers with SSC/HSC. The farmers in the east coast region were mainly primary having education representing 36.9%. The shrimp farmers in Andhra Pradesh state possessed mainly primary education representing 40% where as Tamil Nadu state represented 55% farmers with SSC and HSC.

4.1.1.2 Occupation

Among the shrimp farmers in India, 57.5% were depending aquaculture alone as the main profession; coast-wise they constituted 61.8% of west coast farmers and 55.4% of east coast farmers. Amongst the states Karnataka shrimp farmers were completely practicing aquaculture as the only profession compared to the Andhra Pradesh farmers (53.3%) and Gujarat farmers (80%).

4.1.1.3 Experience

The results indicated that countrywide 65.8% farmers possessed more than 3 years experience. Their composition was more or less equal on the west coast (65.5%) and east coast (64.6%) regions. The percentage of shrimp

Table 4(a). National and Coast-wise profile of characteristics of Shrimp Farmers and Shrimp Farms

S.No	Characteristics*	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1.	Education						
	Illiterate (0)	1	0.8	1	1.5	0	0
	Primary (1)	25	20.8	24	36.9	1	1.8
	SSC/HSC (2)	49	40.8	21	32.3	28	50.9
	Graduation (3)	40	33.3	18	27.7	22	40.0
	Professional (4)	5	4.1	1	15	4	7.3
2.	Occupation						
	Aqua+Agri (1)	26	21.7	17	26.2	9	16.4
	Aqua only (2)	70	58.3	36	55.3	34	61.8
	Aqua+service+ Aqua busi (3)	24	20	12	18.5	12	21.8
3.	Experience						
	No exp (0)	1	0.8	1	1.5	0	0
	One year (1)	4	3.3	2	3.1	2	3.6
	Two years (2)	17	14.2	10	15.4	7	12.7
	Three years (3)	20	16.6	10	15.4	10	18.2
	More than three years (4)	78	65	42	64.6	36	65.5
4.	Farm type						
	Leased (1)	42	35	17	20.0	25	45.5
	Own (2)	78	65	48	80.0	30	54.5
5.	Farm Area (ha)						
	0 – 2 (1)	48	40	26	40	21	38.2
	3 – 5 (2)	45	37.5	25	38.5	20	36.4
	More than 5 (3)	27	22.5	14	21.5	14	25.5
6.	Soil type						
	Sandy loamy (1)	52	43.3	25	38.5	27	49.1
	Sandy clay (2)	48	40	25	38.5	23	41.8
	Clay loam (3)	20	16.6	15	23.1	5	9.1

* Parenthesis indicates scoring; Fr-Frequency

Table 4(b). State wise Profile of Characteristics of Shrimp Farmers and Shrimp Farms

S.No	Characteristics*	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1	Education						
	Illiterate (0)	02.2	00.0	0.00	0.00	0.00	0.00
	Primary (1)	40.0	25.0	06.7	00.0	0.00	0.00
	SSC/HSC (2)	22.2	55.0	53.3	41.7	23.1	80.0
	Graduation (3)	31.1	20.0	20.0	50.0	76.9	20.0
Professional (4)	4.4	0.00	20.0	08.3	00.0	00.0	
2	Occupation						
	Aqua+Agri (1)	37.8	5.0	13.3	00.0	00.0	46.7
	Aqua only (2)	53.3	50.0	80.0	33.3	100.0	33.3
	Aqua+ser+Aqua business (3)	08.9	45.0	06.7	66.7	00.0	20.0
3	Experience						
	No exp (0)	02.2	00.0	00.0	00.0	00.0	00.0
	One year (1)	02.2	50.0	13.3	00.0	00.0	00.0
	Two years (2)	15.6	15.0	13.3	33.3	00.0	6.7
	Three years (3)	11.1	25.0	46.7	00.0	07.7	13.3
More than three years (4)	68.9	55.0	26.7	66.7	92.3	80	
4	Farm Area (ha)						
	0 – 2 (1)	44.4	30.0	26.7	33.3	61.5	40
	3 – 5 (2)	37.8	40.0	53.3	41.7	23.1	26.7
More than 5(3)	17.8	30.0	20.0	25.0	15.4	33.3	
5	Farm type						
	Leased (1)	33.3	15.0	86.7	25.0	15.4	46.7
Own (2)	66.7	85.0	13.3	75.0	84.6	53.3	
6.	Soil type						
	Sandy loamy (1)	37.8	35.0	20.0	25.0	53.8	93.3
	Sandy clay (2)	33.3	50.0	80.0	41.7	38.5	06.7
Clay loam (3)	28.9	15.0	00.0	33.3	07.7	00.0	

* Parenthesis indicates scoring

farmers with experience of more than three years was high in Andhra Pradesh (68.9%) and Karnataka (80%).

4.1.1.4 Farm Type

Farmers who were performing shrimp culture in their own farms were 65% country-wide, and coast-wise, 80% farmers were from east coast region and only 54.5% in the west coast region; the rest were taking up shrimp culture in leased farms. Amongst the states, farmers from Andhra Pradesh and Karnataka states were doing the shrimp culture in their own farms whereas on the other end of the spectrum 86.7% farmers from Gujarat state were doing shrimp culture in leased ponds.

4.1.1.5 Soil Type

The results revealed that 43.3% of Indian shrimp farmers were taking up shrimp culture in sandy loamy type soils and 40% in sandy clay soils. On the east coast region, 38.5% of shrimp farmers were using sandy loamy and sandy clay type soils, and on the west coast, 49.1% of sandy loamy soils. Amongst the states Gujarat state farmers were doing shrimp culture in sandy clay type soils, where as in Andhra Pradesh 50% shrimp farms were on sandy clay type soil. In Maharashtra the farms that were having clay loamy type soil represented 33.3%.

4.1.1.6 Farm Area

The results of the present survey revealed that shrimp farmers who were holding 0-2 ha of farm area constituted 40% of the total farmers in the region. Amongst the states, 44.4% farmers from Andhra Pradesh and 61.5% from Karnataka state were holding 0.2 ha. The farmers from Tamil Nadu (40%) and Gujarat (53.3%) states were having higher land holdings of 3-5 ha.

4.1.2 Characteristics of Intensity of Shrimp Farming

The results pertaining to intensity of shrimp farming are presented in Tables 5(a) and 5(b).

Table 5(a). National and Coast-wise intensity of Shrimp Farming

S.No	Characteristics	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1.	Number of crops						
	One (1)	17	14.2	13	20	4	07.3
	Two (2)	92	76.7	52	80	40	072.7
	Three (3)	11	09.2	0	0	11	20.0
2.	Crop rotation						
	Yes (1)	18	15.0	17	26.2	01	01.8
	No (2)	102	85.0	48	73.8	54	98.2
3.	Aerators						
	No (0)	34	28.3	13	20.0	20	36.4
	Yes (1)	86	71.7	52	80.0	35	63.6
4.	Culture period (Days)						
	Below 120 (1)	51	42.5	24	36.9	27	49.1
	120-150 (2)	69	57.5	41	63.1	28	50.9
5.	Water source						
	Bore well (1)	32	26.7	29	44.6	3	5.5
	Creek (2)	88	73.3	36	55.4	52	94.5
6.	Stocking density (/m²)						
	More than 15 (1)	11	09.2	9	13.8	2	03.6
	13-15 (2)	28	23.3	21	32.3	7	12.7
	8-12 (3)	38	31.7	10	15.4	28	50.9
	5-7 (4)	43	35.8	25	38.5	18	32.7
7.	Water exchange						
	No exchange (0).	29	24.2	10	15.4	19	34.5
	Every 30days (1)	27	22.5	22	33.8	5	09.1
	15days (2)	37	30.8	12	18.5	25	45.5
	7 days (3)	27	22.5	21	32.3	6	10.9
8.	Water depth (m)						
	1-1.5 (1)	10	08.3	4	06.2	6	10.9
	Less than1 (2)	110	91.7	61	93.8	49	89.1
9	Months of stocking						
	Jan/Feb/March 2)	102	85	48	73.8	54	98.2
	June/July/Aug (1)	102	85	48	73.8	54	98.2

* Parenthesis indicates scoring ; Fr-Frequency

Table 5(b). State wise- Intensity of shrimp farming

S.No	Characteristics	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1	No of crops						
	One (1)	26.7	05.0	06.7	25.0	00.0	00.0
	Two (2)	73.3	95.0	93.3	75.0	100	26.7
	Three (3)	00.0	00.0	00.0	00.0	00.0	73.3
2	Crop rotation						
	Yes (1)	30.1	00.0	00.0	00.0	00.0	06.7
	No (2)	68.9	95.0	100	100	100	93.3
3	Aerators						
	No (0)	28.9	00.0	13.3	16.7	07.7	100
4	Yes (1)	71.1	100	86.7	83.3	92.3	00.0
	Culture period (days)						
	< 120 (1)	44.4	20.0	06.7	25.0	69.2	93.3
5	120-150 (2)	55.6	80.0	93.3	75.0	30.8	06.7
	Water source						
6	Bore well (1)	60.0	05.0	13.3	08.3	00.0	00.0
	Creek (2)	40.0	95.0	86.7	91.7	100	100
7	Stocking density (No. of PL/m²)						
	<15 (1)	11.1	15.0	00.0	00.0	07.7	06.7
	13-15 (2)	11.1	80.0	00.0	16.7	38.5	00.0
	8-12 (3)	22.2	00.0	66.7	33.3	53.8	46.7
	5-7 (4)	55.6	05.0	33.3	50.0	00.0	46.7
8	Water exchange						
	No Exchange (0)	20.0	05.0	00.0	16.7	30.8	93.3
	Every 30 Days (1)	22.2	50.0	06.7	00.0	30.8	00.0
	15Days (2)	26.7	10.0	66.7	91.7	30.8	00.0
	7 Days (3)	31.1	35.0	26.7	00.0	7.7	06.7
9	Water depth (cm)						
	1-1.5 (1)	08.9	00.0	00.0	50.0	00.0	00.0
	< 1 (2)	91.1	100.	100	50.0	100	100
9	Months of stocking						
	Jan/Feb/March (2)	68.9	95	100	100	100	93.3
	June/July/Aug (1)	68.9	95	100	100	100	93.3

* Parenthesis indicates scoring ; Fr - Frequency

4.1.2.1 Stocking Density

35.8% farmers all over India were stocking 5-7 number of post larvae per square meter whereas 31.7% farmers were stocking 8-12 number of post larvae per square meter. Region wise, 38.5% farmers from east coast were stocking 5-7 number of post larvae per square meter whereas 32.3% farmers were stocking 13-15 number of post larvae per square meter. Majority (50.9%) of farmers from west coast region were stocking 8-12 number of post larvae per square meter. Amongst the states, 80% of farmers from Andhra Pradesh were stocking 13-15 number of post larvae per square meter and 66.7% farmers from Gujarat were stocking 8-12 number of post larvae per square meter.

4.1.2.2 Months of Stocking

Majority of farmers from all over India were stocking the seed during the months of January/February for the first crop and June/July for the next crop. Region wise 73.8% of farmers from east coast and 98.2% farmers from west coast were stocking the shrimp seed during the months of January/February for first crop and June/July for the next crop. Amongst the states all (100%) farmers from the states of Gujarat, Maharashtra and Karnataka, 68.9% of farmers from Andhra Pradesh and 95% of farmers from Tamil Nadu were stocking during the months of January/February for the first crop and June/July for the second crop.

4.1.2.3 Culture Period

Majority (69%) of the farmers all over India were maintaining the culture period of 120-150 days. Region wise 63.1% of farmers from east coast and 50.9% of farmers from west coast region were maintaining the culture period of 120-150 days. Amongst the states, 80% of farmers were maintaining the culture period of 120-150 days where as 69.2% of farmers from Karnataka and 93.3% farmers from Kerala were maintaining the culture period of 120-150 days.

4.1.2.4 Number of Crops per Year

76.7% farmers all over India were practicing two crops per year. Region-wise 80% farmers in the east coast region and 72.7% farmers in the west coast region were practicing two crops per year. 20% farmers in the west coast region were mainly Kerala practicing three crops per year. 100% of the Karnataka farmers were taking two crops per year and 95% and 73.3% of farmers respectively from Tamil Nadu and Andhra Pradesh were taking two crops per year.

4.1.2.5 Crop Rotation

Most of the Indian shrimp farmers (85%) were practising exclusively shrimp farming alone in their farms and few (15%) of the farmers were performing crop rotation either with paddy or fish culture. Crop rotation was mostly observed in the east coast (26%) shrimp farms. Amongst the states, Andhra Pradesh (30%) and Kerala (67%) state farmers resorted to the practices of crop rotation.

4.1.2.6 Aerators

The results revealed that 86% of the Indian shrimp farmers were using aerators (Plate1) and the usage was more prevalent in the east coast shrimp farms (80%) than in west coast (63.6). Amongst the states, 100% of farmers from Tamil Nadu were using aerators whereas 71.1% Andhra Pradesh farmers had aerators in their farms. None of the Kerala shrimp farmers in the present study were using any aerators.

4.1.2.7 Water Exchange

The results indicated that 30.8% of the shrimp farmers in India were carrying water exchange for every 15 days and it was more commonly (45.5%) carried by west coast farmers. The results also indicated that almost $\frac{3}{4}$ th of the farmers were carrying water exchange and only 15.4% of farmers on the east coast and 34.5% on the west coast were not doing water exchange. Amongst the states 50% of Tami Nadu state farmers were carrying

water exchange once every 30 days where as Andhra Pradesh farmers were more or less equally divided over all the four categories of water exchange. Amongst the Karnataka farmers 30.8% were carrying out water exchange equally for every 30 days and 15 days and 30.8% farmers were not at all carrying out any water exchange.

4.1.2.8 Water Depth

Majority of the shrimp farmers (91.7%) all over India were maintaining optimal water depth, which is essential for cultivable shrimps, and there was not much variations amongst the states of east coast and west coast regions.

4.1.2.9 Water Source

Majority (73.3%) of the shrimp farmers in India were using creek water from creeks as the water source for the shrimp farms. Most of the west coast farmers (94.5%) were using water from creeks for their shrimp farms compared to east coast farmers (55.4%). 60% of the farmers from Andhra Pradesh were using ground water as the main water source.

4.1.3 Shrimp Farm Management Practices

The results of the present study regarding the management practices followed in various shrimp farms are presented in Tables 6(a) and 6(b).

4.1.3.1 pH

The results indicated that most of the farmers (99.2%) were maintaining the pH at 7.8 to 8.5 (Plate 2).

4.1.3.2 Salinity

The results indicated that 60% of the farmers were practicing shrimp farming either 5-15 ppt or 25-30 ppt and the remaining 40% of the farmers only maintaining optimal salinity of 15 to 25 ppt. In west coast about half of the farmers were maintaining both the salinities. Amongst the east coast 73.3% of the shrimp farmers were culturing at 5-15 ppt salinities and Tamil Nadu farmers were culturing at both the salinities.

Table 6(a). National and Coast-wise Water Quality Management Practices in Shrimp Farming

S. No	Parameter	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1	pH 6 – 7.5 (or) 8.5 – 11 (1) 7.5 – 8.5 (2)	1	00.8	0	00.0	1	01.8
		119	99.2	65	100	54	98.2
2	Salinity (ppt) 5 – 15 (or) 25 – 30 (1) 15 – 25 (2)	72	60.0	44	67.5	28	50.9
		48	40.0	21	32.3	27	49.1
3	Temperature (°c) < 28 and >32 (1) 28 – 32 (2)	25	20.8	23	35.4	2	03.6
		95	79.2	42	64.6	53	96.4
4	Transparency (cm) <30 or >40 (1) 30 – 40 (2)	25	20.8	15	23.1	10	18.2
		95	79.2	50	76.9	45	81.8
5	DO (ppm) Less than 3 (0) 3 – 5 (1) More than 5 (2)	87	72.5	55	84.6	32	58.2
		10	08.3	0	00.0	10	18.2
		23	19.2	10	15.5	13	23.6
6	Plankton Poor (1) Medium (2) Rich (3)	87	72.5	46	70.8	41	74.5
		30	25.0	19	29.2	11	20.0
		3	02.5	0	00.0	3	05.5
7	Water color Other than (1) Light green (2)	25	20.8	15	23.1	10	18.2
		95	79.2	50	76.9	45	81.8
8	Frequency of Testing No tests (0) 30 Days (1) 15 Days (2) 7 Days (3)	2	01.7	0	00.0	2	03.6
		65	54.2	40	61.5	25	45.5
		43	35.8	16	24.6	27	49.1
		10	08.3	9	13.8	1	01.8

* Parenthesis indicates scoring ; Fr- Frequency

Table 6(b). State Wise Water quality Management Practices in Shrimp Farming

S.No	Parameter	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1.	pH						
	6 – 7.5 or 8.5 – 11 (1)	100	100	100	100	100	6.7
2.	7.5 – 8.5 (2)	00.0	00.0	00.0	00.0	00.0	93.3
	Salinity (ppt)						
3.	5 – 15 or 25 – 30 (1)	73.3	50.0	13.3	91.7	7.7	93.3
	15 – 25 (2)	26.7	50.0	86.7	8.3	92.3	6.7
3.	Temperature (°c)						
	< 28 and >32 (1)	44.4	0.05	06.7	100	100	06.7
4.	28 – 32 (2)	55.6	95.0	93.3	00.0	00.0	93.3
	Transparency (cm)						
5.	<30 or >40 (1)	28.9	100	06.7	75.0	100	100
	30 – 40 (2)	71.1	0	93.3	25.0	00.0	00.0
5.	DO (ppm)						
	Less than 3 (0)	80	100	26.7	100	7.7	100
6.	3 – 5 (1)	20	00.0	73.3	00.0	76.9	00.0
	More than 5 (2)	0	00.0	00.0	00.0	15.4	00.0
6.	Plankton						
	Poor (1)	84.4	35.0	33.3	75.0	100	93.3
7.	Medium (2)	15.6	65.0	60.0	08.3	00.0	06.0
	Rich (3)	00.0	00.0	6.7	16.7	00.0	0.0
7.	Water color						
	Other than (1)	22.2	30.0	06.7	75.0	100	100
8.	Light green (2)	77.8	70.0	93.3	25.0	00.0	00.0
	Frequency of Testing						
8.	No tests (0)	02.0	01.7	00.0	00.0	02.0	03.6
	30 Days (1)	66.7	45.0	13.3	58.3	23.1	13.3
9.	15 Days (2)	17.8	45.0	86.7	33.3	76.9	86.7
	7 Days (3)	15.6	10.0	00.0	08.3	00.0	00.0

* Parenthesis indicates scoring

4.1.3.3 Transparency

Most of the shrimp farms (79.2%) in India were having the optimal turbidity. 93.3% of the farms in Gujarat and 71.1% of farms in Andhra Pradesh were having the optimal turbidity of 30-40 cm. All the farmers of Tamil Nadu, Karnataka and Kerala maintain the transparency at optimal levels.

4.1.3.4 Dissolved Oxygen (DO)

It is interesting to observe that 72.5% of the Indian shrimp farmers were maintaining dissolved oxygen (DO) levels around 3 ppm in their farms. Region-wise, 84.6% farms on the east coast 58.2% on the west coast region were having DO levels of 3 ppm. All the farmers of Tamil Nadu, Maharashtra, and Kerala and 80% farmers in Andhra Pradesh were maintaining DO levels at 3 ppm.

4.1.3.5 Plankton

The results showed that 72.5% of the Indian shrimp farms were having poor plankton growth. Mainly Karnataka (100%) and Andhra Pradesh (84.4%) farms were having the poor plankton growth compared to Gujarat where majority (73.3%) of the shrimp farms were having medium plankton growth.

4.1.3.6 Water Colour

It is interesting to observe that the most (95%) of the farmers were maintaining green brown or yellow brown (optimal colour as suggested by MPEDA, 1996) in spite of poor plankton growth. The results were more or less similar to the east coast (76.9%) and west coast (81.8%) region. All the shrimp farms (100%) of Karnataka and Kerala were reporting below optimal water colour. The farmers of Gujarat, Karnataka and Kerala were have more similar results with that of plankton presence and water colour.

4.1.3.7 Frequency of Testing Water Quality Parameters

More than half of the farmers (54.2%) in India were testing the water quality parameters (Plate 3) every month, and only negligible (1.7%) number



Plate 1: Showing a paddle wheel aerator in a shrimp culture pond



Plate 2: Showing application of lime in a shrimp culture pond



Plate 3: Showing preparation for biological sampling in a shrimp culture pond

of farmers were not at all testing any of these parameters. East coast shrimp farmers (61.5%) were testing these parameters every month whereas west coast shrimp farmers were testing them every month (45.5%) and every fortnight(49.1%).Most of the Andhra Pradesh farmers (66.7%) were testing these parameters for every month and farmers of Gujarat (86.7%) and Karala (86.7%) were testing these parameters every fortnight.

4.1.4 Characteristics of Seed and Feed

The results pertaining to the seed and feed sources were presented in Tables 7(a) and 7(b).

4.1.4.1 Source of Seed

The results showed all the Indian shrimp farmers (100%) were using hatchery produced post-larvae.

4.1.4.2 Age of PL

Majority (88.3%) of the shrimp farmers were stocking 15-20 days of post-larvae. There was not much difference between the practices adopted by east coast and west coast region farmers.

4.1.4.3 Seed Testing

The survey results indicated that about half of the Indian shrimp farmers (48.3%) were performing PCR testing (Plate 4) prior to stocking in grow-out ponds. Majority (60%) of the west coast farmers were carrying out PCR test for their post larvae, while 47.6% of the east coast farmers carrying all tests apart from PCR testing. A few Indian farmers (10%) were not carrying out any test for the post larvae prior to purchasing them from the hatchery. Majority of Gujarat (86.6%) and Kerala (86.6%) farmers were performing the PCR test for the hatchery seed. Majority of Andhra Pradesh farmers were conducting all the tests (31.1%) and PCR test (48.8%) which adds upto an overall 79.9% farmers who conduct essential tests for seed quality

Table 7(a). National and Coast-wise Characteristics of Sources of Seed and Feed used in Shrimp Farming

S.No	Characteristics	India		East-coast		West coast	
		Fr	%	Fr	%	Fr	%
1.	Feed type						
	Local (1)	2	01.7	1	01.5	1	01.8
	Local + branded (2)	15	12.5	1	01.5	14	25.5
	Only branded (3)	103	85.8	63	96.9	40	72.7
2.	FCR						
	>1.8 (1)	17	14.2	12	18.5	5	09.1
	1.6 – 1.8 (2)	34	28.3	13	20.0	21	38.2
	<1.5 (3)	69	57.5	40	61.5	29	52.7
3.	Source of PL						
	Wild (1)	0	00.0	0	00.0	0	00.0
	Hatchery (2)	120	100	65	100	55	100
4.	Age of PL (days)						
	More than 20 (1)	14	11.7	8	12.3	6	10.9
	15-20 (2)	106	88.3	57	87.2	49	89.1
5.	Seed testing						
	No test (0)	12	10.0	7	10.7	5	09.09
	Stress test (1)	3	02.5	2	03.6	1	01.8
	PCR test (2)	58	48.3	25	38.4	33	60.0
	All tests (3)	47	39.1	31	47.6	16	29.0

* Parenthesis indicates scoring
Fr=Frequency

Table 7(b). State wise Characteristics of Seed and Feed used in Shrimp Farming

S.No	Characteristics	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1.	Source of Feed						
	Local (1)	2.2	5	100	100	7.7	6.7
	Local & branded (2)	97.8	95	0	0	92.3	86.7
	Only branded (3)	0	0	0	0	0	6.7
2.	FCR						
	>1.8 (1)	13.3	30	6.7	33.3	100	6.7
	1.6 – 1.8(2)	20	20	93.3	58.3	0	86.7
	<1.5 (3)	66.7	50	0	8.3	0	6.7
3.	Source of seed						
	Wild (1)	0	0	0	0	0	0
	Hatchery (2)	100	100	100	100	100	100
4.	Age of PL						
	> 20 (1)	17.8	0	6.7	8.3	7.7	20
	15-20 (2)	82.2	100	93.3	91.7	92.3	80
5.	Seed Testing						
	No test (0)	15.5	0	0	2.5	7.6	6.6
	Stress test (1)	4.4	0	6.6	0	0	0
	PCR test (2)	48.8	25	86.6	41.6	7.6	86.6
	All tests (3)	31.1	75	6.6	33.3	84.6	6.6

* Parenthesis indicates scoring



Plate 4: Showing PCR maintained in a shrimp culture farm in AP



Plate 5: Showing shrimp feed stored in a dealer's godown



Plate 6: Showing feed being broadcast in a shrimp culture pond

4.1.4.4 Source of Feed

Majority (85.8%) of the Indian shrimp farmers were using branded feed (Plate 5) alone as the feed source. Almost all (96.9%) east coast farmers were using only the commercial feed but in Kerala, majority of farmers (86.7%) were using both local / farm made feeds and branded feeds. Plates 6 and 7 show the feed dispersal in the ponds.

4.1.4.5 Feed Conversion Ratio (FCR)

More than half of the farmers (57.5%) in India were maintaining FCR values below 1.5%. All the farmers in Karnataka (100%) and majority of the Gujarat farmers (93.3%) were maintaining FCR of less than 1.5%.

4.1.5 Chemicals used in Shrimp Farming

The results pertaining to the usage of antibiotics, molt inducers, sanitizers and probiotics are presented in Tables 8(a) and 8(b).

4.1.5.1 Antibiotics

66.7% of the shrimp farmers in India did not respond when questioned about the usage of antibiotics; 30.8% informed that they were not using any antibiotics and only very few (2.5%) farmers confessed about the usage of antibiotics (Plates 8-10). In the east coast region 70.8% of the shrimp farmers did not respond to queries on antibiotic usage; majority (86.7%) of the shrimp farmers from Andhra Pradesh and all (100%) farmers from Tamil Nadu did not respond to queries on usage of antibiotics. 4.4% of the Andhra Pradesh farmers informed about the usage of antibiotics and 8.9% farmers answered in the negative. The results are presented in Fig. 2.

4.1.5.2 Molt Inducers

The results indicated that 45.8% of the shrimp farmers in India were using molt inducers and there was not much difference between the east coast (49.2%) and west coast (42.8%) regions. 47.5% of the shrimp farmers in India did not respond to queries about the usage of molt inducers 55% of

Table.8 (a) National and Coast-wise Usage of Antibiotics and Chemicals in Shrimp Farming

S.No	Chemicals	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1.	Antibiotics						
	Yes (2)	3	2.5	2	3.1	1	1.8
	No (1)	37	30.8	17	26.2	20	36.4
	NR (0)	80	66.7	46	70.8	34	61.8
2.	Molt inducers						
	Yes (2)	55	45.8	32	49.2	23	41.8
	No (1)	8	6.7	4	6.2	4	7.3
	NR (0)	57	47.5	29	44.6	28	50.9
3.	Sanitizers						
	Yes (2)	57	47.5	32	49.2	25	45.5
	No (1)	2	1.7	2	3.1	0	0
	NR (0)	61	50.8	31	47.7	30	54.5
4.	Probiotics						
	Yes (2)	90	75.0	51	78.5	39	70.9
	No (1)	13	10.8	11	16.9	2	3.6
	NR (0)	17	14.2	3	4.6	14	25.5

* Parenthesis indicates scoring ; Fr- Frequency

** NR - No Response

Table 8(b). State wise Usage of Antibiotics and Chemicals in shrimp farming

S.No	Chemicals	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1.	Antibiotics						
	Yes (2)	04.4	00.0	26.7	16.7	00.0	06.7
	No (1)	08.9	65.0	73.3	00.0	100	06.7
	NR (0)	86.7	35.0	00.0	83.3	00.0	86.7
2.	Molt Inducers						
	Yes (2)	00.0	20.0	0	16.7	15.4	00.0
	No (1)	51.1	55.0	100	66.7	00.0	00.0
	NR (0)	48.9	25.0	00.0	16.7	84.6	100
3.	Sanitizers						
	Yes (2)	00.0	10.0	100	00.0	07.7	00.0
	NO (1)	40.0	80.0	00.0	75.0	00.0	00.0
	NR (0)	60.0	10.0	00.0	25.0	92.3	100
4.	Probiotics						
	Yes (2)	68.9	100	100	83.3	100	06.7
	No (1)	24.4	00.0	00.0	16.7	00.0	00.0
	NR (0)	06.7	00.0	00.0	00.0	00.0	93.3

* Parenthesis indicates scoring

** NR - No Response

Fig. 2. Usage of Antibiotics in Indian Shrimp Farming

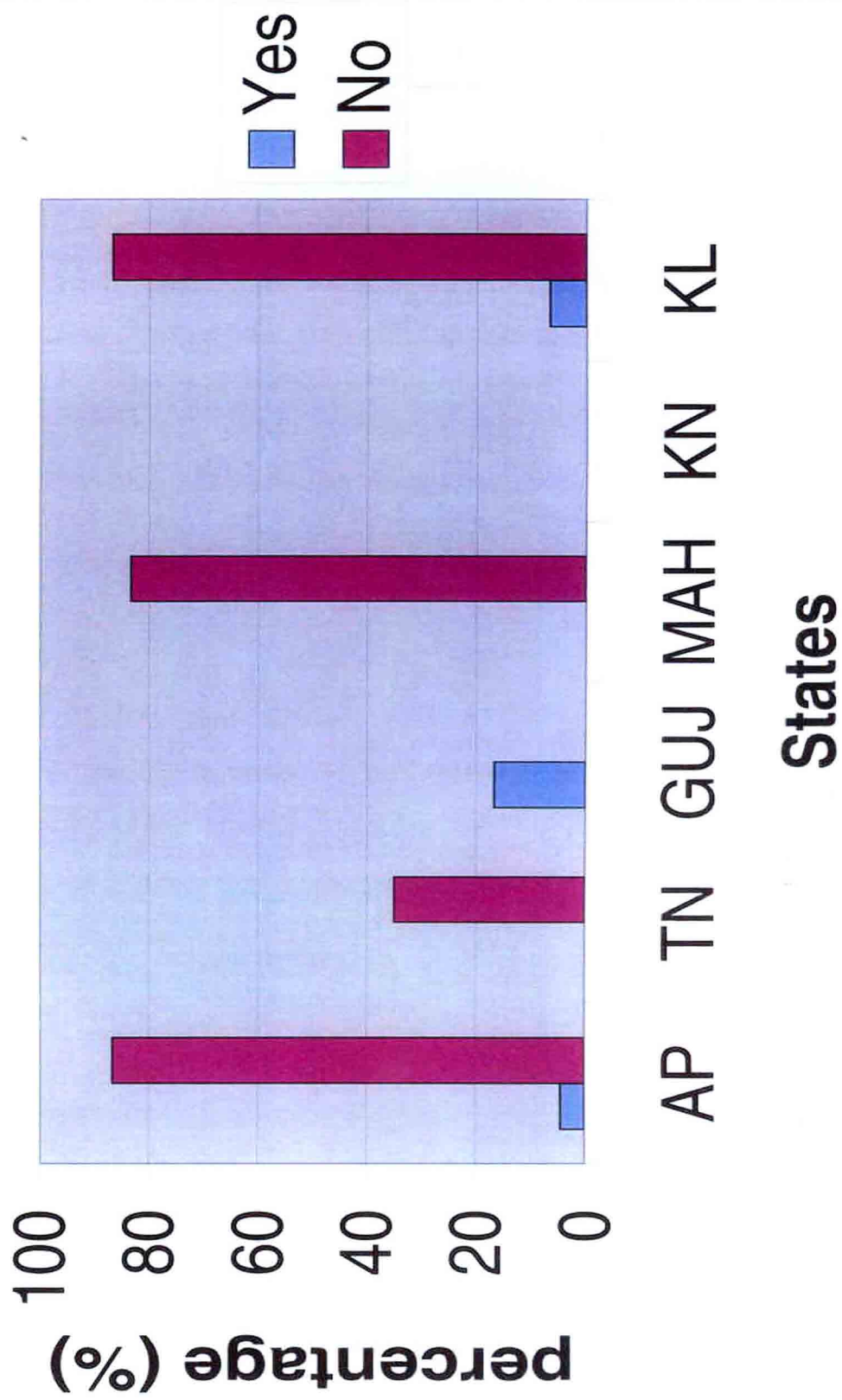




Plate 7: Showing feed being broadcast in a shrimp culture pond



Plate 8: Showing on-farm preparation of feed mixed with antibiotic



Plate 9: Showing on-farm preparation of feed mixed with antibiotic

Tamil Nadu and 51.1% of the Andhra Pradesh farmers were using molt inducers in their shrimp culture practices. 73.3% of farmers from Gujarat, and 66.7% of farmers from Maharashtra were not using any molt inducers.

4.1.5.3 Sanitizers

The results showed that 47.5% of the shrimp farmers in India were using the sanitizers; region-wise they constitute 49.2% of east coast and 45.5% of west coast farmers. 80% of the Tamil Nadu farmers and 51.1% of the Andhra Pradesh farmers were not using any sanitizers.

4.1.5.4 Probiotics

Majority (75%) of the Indian shrimp farmers were using the probiotics in their farm management practices (Fig. 3). A large number of farmers on east coast (78.5%) and west coast (70.9%) were using probiotics. Amongst the states all (100%) the farmers of Tamil Nadu, Gujarat and Karnataka were using the probiotics. The farmers in Andhra Pradesh (68.9%) and Maharashtra (83.3%) were using reasonable amount of probiotics in their farming practices. 93.3% of the shrimp farmers in Kerala did not respond to the queries on the usage of probiotics and only 6.7% of the farmers confessed about the usage of probiotics.

4.1.6 Characteristics of Marketing

The results pertaining to characteristics of marketing were presented in Tables 9(a) and 9(b).

The results indicated that 58.3% of the Indian shrimp farmers were selling their harvested (Plates 11 and 12) raw material directly to export firms whereas 41.7% the shrimp farmers were selling them through agents. Majority (90.8%) of the shrimp farmers were selling the raw material as 'Head-on' type. All (100%) farmers of Tamil Nadu and majority (93.3%) of the Kerala farmers were selling their raw material directly to the export firms whereas 60% of the farmers from Andhra Pradesh and all the farmers of Karnataka were selling them through agents or middle men.

Table 9(a). National and Coast-wise Profile of Characteristics of Marketing

S.No	Characteristics	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1.	Marketing						
	Agent (1)	50	41.7	28	43.1	22	40.0
	Company (2)	70	58.3	37	56.9	33	60.0
2.	Raw material						
	Head less (1)	11	09.2	8	12.3	3	05.5
	Head on (2)	109	90.8	57	87.7	52	94.5

* Parenthesis indicates scoring ; Fr - Frequency

Table 9(b). State wise Profile of characteristics of Marketing

S.No	CHARACTERS	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1.	Marketing						
	Agent (1)	60.0	00.0	13.3	50.0	100	06.7
	Company (2)	40.0	100	86.7	50.0	00.0	93.3
2.	Raw material						
	Head less (1)	17.8	00.0	06.7	16.7	00.0	100
	Head on (2)	82.2	100	93.3	83.3	100	00.0

* Parenthesis indicates scoring ; Fr-Frequency

Fig.3.Usage of Antibiotics and Probiotics in Indian Shrimp Farming

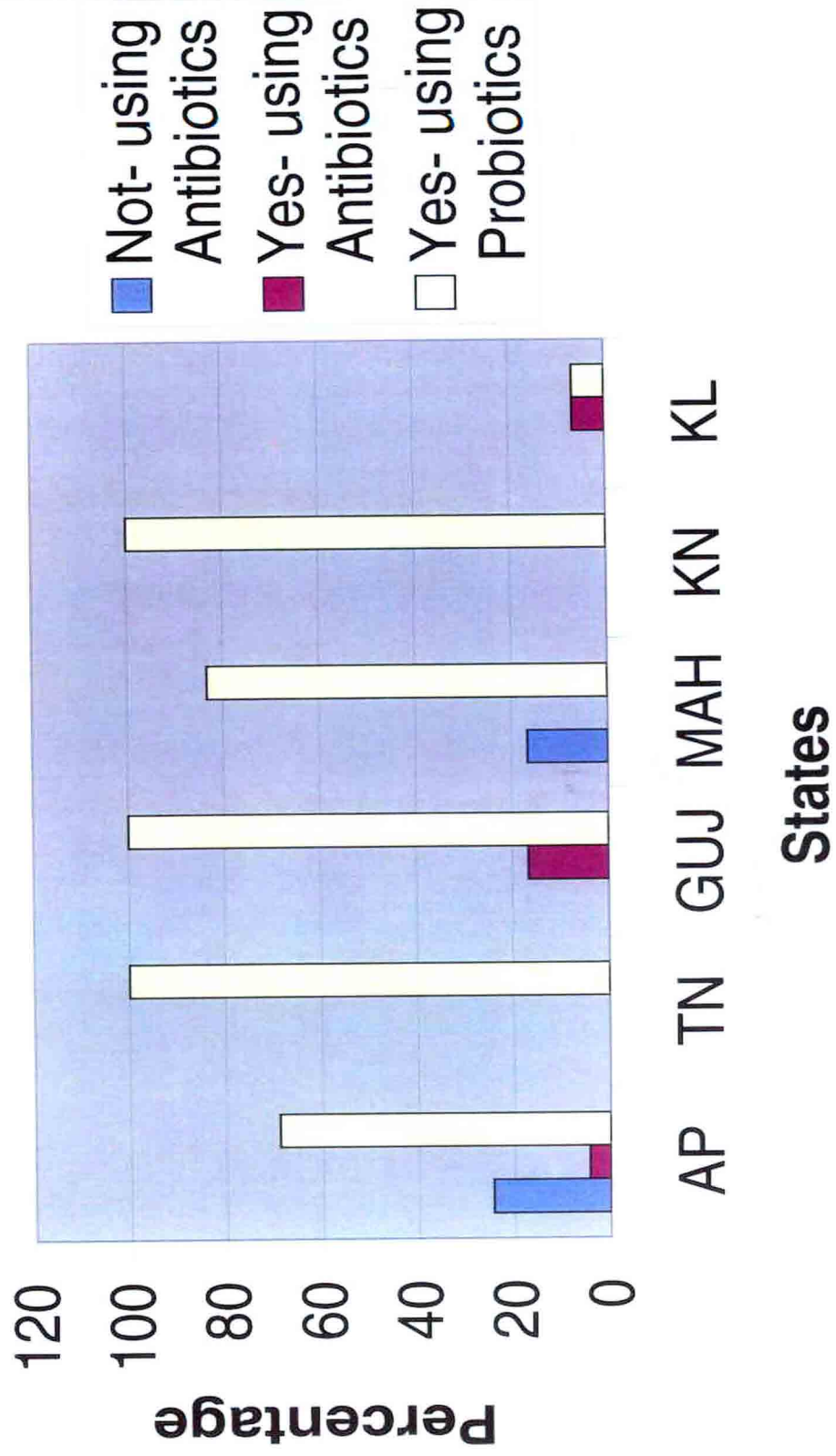




Plate 10: Showing the drying of prepared feed mixed with antibiotic



Plate 11: Showing harvesting of shrimp by cast net



Plate 12: Showing harvesting of shrimp by drag net

4.1.7 Technical Awareness Related to Shrimp Farming

The results pertaining to the technical awareness on aspects related to the shrimp farming are presented in Tables 10(a) and 10(b).

4.1.7.1 Training Experience

The results indicated that 71.7% of the Indian shrimp farmers did not have any prior training on aspects related to shrimp farming. 80% of east coast farmers were doing the shrimp farming with their own experience compared to 61.8% farmers on the west coast. 93.3% farmers of Gujarat and 50% of farmers of Maharashtra had undergone training on aspects related to shrimp farming.

4.1.7.2 Willingness to Undergo Training

Majority (76.7%) of the total Indian shrimp farmers had evinced interest to undergo training in advances of shrimp farming technology but 69.2% farmers from Karnataka and 41.7% farmers Maharashtra were not interested to undergo any training.

4.1.7.3 Awareness of Banned Antibiotics

Majority (88.3%) of the Indian shrimp farmers were aware of banned antibiotics and both the coasts yielded similar results with 87.7% farmers of east coast and 89% farmers of west coast being aware of the ban. All the farmers (100%) of Tamil Nadu and Gujarat states were aware about banned antibiotics. Only 17.8% of the Andhra Pradesh and 26.7% of the Kerala farmers were not aware of banned antibiotics.

4.1.7.4 Willingness to Adopt New Technology

Positive results were obtained with respect to the farmer's willingness to accept new technologies. Majority (86.7%) of the shrimp farmers in India expressed their interest to accept any newly developed technology to improve the quality of the raw material. Only Gujarat farmers (66.7%) had shown their

Table 10(a). National and Coast-wise Characteristics of Technical Awareness related to Shrimp Farming

S. No	Characteristics	India		East-coast		West-coast	
		Fr	%	Fr	%	Fr	%
1	Training exp						
	No (0)	86	71.7	52	80.0	34	61.8
	Yes (1)	34	28.3	13	20.0	21	38.2
2	Training willingness						
	No (0)	28	23.3	10	15.4	18	32.7
	Yes (1)	92	76.7	55	84.6	37	67.3
3	Awareness of banned antibiotics						
	No (0)	14	11.7	8	12.3	06	10.9
	Yes (1)	106	88.3	57	87.7	49	89.1
4	Willingness to adopt to new technology						
	No (0)	16	13.3	04	06.2	12	21.1
	Yes (1)	104	86.7	61	93.8	43	78.2
5	Awareness of MPEDA guidelines						
	No (0)	58	48.3	32	49.2	26	47.3
	Yes (1)	62	51.7	33	50.8	29	52.7
6	Source of Technical help						
	Fellow farmer / self (1)	32	26.7	18	27.7	14	25.5
	Technician (2)	74	61.7	44	67.7	30	54.5
	Professionals (3)	14	11.7	3	04.6	11	20.0

* Parenthesis indicates scoring ; Fr-Frequency

Table 10(b). State wise Characteristics of Technical Awareness in Shrimp Farming

S.No	Characters	States (%)					
		A.P	TN	GUJ	MAH	KN	KL
1.	Undergone Training						
	No (0)	73.3	90.0	06.7	50.0	100	93.3
	Yes (1)	26.7	10.0	93.3	50.0	00.0	06.7
2.	Willingness to Training						
	No (0)	15.6	10.0	06.7	41.7	69.2	20.0
	Yes (1)	84.4	90.0	93.3	58.3	30.8	80.0
3.	Awaroness of Banned Antibiotics						
	No (0)	17.8	00.0	06.7	00.0	07.7	26.7
	Yes (1)	82.2	100	93.3	100	92.3	73.3
4.	Willingness to New Technology						
	No (0)	11.1	00.0	66.7	16.7	00.0	00.0
	Yes (1)	88.9	100	33.3	83.3	100	100
5.	Awareness of MPEDA Guidelines						
	No (0)	53.3	45.0	66.7	16.7	15.4	80.0
	Yes (1)	46.7	55.0	33.3	83.3	84.6	20.0
6.	Source of Technical Help						
	Fellow farmer/self (1)	40.0	05.0	73.3	00.0	15.4	06.7
	Technician (2)	53.3	95.0	13.3	66.7	84.6	60.0
	Professional (3)	06.7	00.0	13.3	33.3	00.0	33.3

* Parenthesis indicates scoring ; Fr-Frequency

unwillingness to adopt any new technology, as they were more comfortable with existing results with respect to the production of quality raw material.

4.1.7.5 Awareness of MPEDA Guidelines

Only 51.7% of the shrimp farmers in India were aware of the MPEDA guidelines and, indicating that nearly half (48.3%) of the farmers in India were not aware any guidelines for improved culture practices. Majority of the Kerala (80%), Gujarat (66.7%) and almost half (45%) of shrimp farmers in Andhra Pradesh were not aware of MPEDA guidelines.

4.1.7.6 Source of Technical Help

Most of the shrimp farmers in India (61.7%) were depending on feed technicians for technical help required during their regular farming practices. In this east coast, a large number of farmers (67.7%) were depending on feed technicians compared to west coast (54.5%) farmers. 73.7% farmers of Gujarat and 66.7% of Maharashtra farmers were depending on their own technologies gained due to the several years of experience in shrimp farming and by discussing with fellow farmers. In Kerala, 33.3% of the shrimp farmers were taking technical help from the professionally qualified people mainly from the Government institutes.

4.1.8 Diseases in shrimp farming

The results pertaining to the diseases caused by viral, bacterial, fungal and protozoan were present in the Tables 11(a) and 11(b).

4.1.8.1 Viral Diseases

36.6% of the viral diseases reported in India was due to White Spot Syndrome Virus (WSSV). Majority (85%) of the Tamil Nadu and 40% of Andhra Pradesh farms were affected due to viral diseases of farmed shrimp but and Gujarat was completely free from viral disease of shrimps.

Table 11(a). National and Coast-wise Problems of Diseases in Shrimp Farming

S.No	Problems	India		East coast		West coast	
		Fr	%	Fr	%	Fr	%
1	Viral	44	36.6	37	56.9	7	12.7
2	Bacterial	29	24.1	25	38.4	4	07.2
3	Fungal	4	03.3	3	04.1	1	01.8
4	Protozoan	12	10.0	1	01.5	11	20.0
5	Others						
	a) Loose Shell	22	18.3	15	23.07	7	12.7
	b) Gill disease	5	04.1	5	07.6	0	00.0

* Parenthesis indicates scoring; Fr-Frequency

Table 11(b). State wise Problems of Diseases in Shrimp Farming

S.No	Problems	States (%)					
		AP	TN	GUJ	MAH	KN	KL
1	Viral	40.0	85.0	00.0	16.6	5.8	20.0
2	Bacterial	42.2	35.0	00.0	00.0	8.8	06.6
3	Fungal	06.6	00.0	00.0	00.0	2.9	00.0
4	Protozoan	02.2	00.0	00.0	00.0	29.4	06.6
5	Others						
	a) Loose Shell	13.3	04.5	00.0	00.0	53.8	00.0
	b) Gill disease	02.2	20.0	00.0	00.0	00.0	00.0

* Parenthesis indicates scoring

4.1.8.2 Bacterial Diseases

24.1% of the diseases of farmed shrimp were caused due to the bacterial infections. Andhra Pradesh (42.2%) and Tamil Nadu (30%) state farmers reported higher levels of bacterial diseases compared to other Indian states. Gujarat and Maharashtra state farmers did not report any bacterial disease.

4.1.8.3 Fungal Diseases

There were very few (4.1%) reported incidents of fungal diseases. Only Andhra Pradesh (6.6%) and Karnataka (2.9%) farmers reported fungal the diseases of farmed shrimps.

4.1.8.4 Protozoan Diseases

Protozoan diseases were reported by 10% of Indian shrimp farmers. Majority of the protozoan diseases were reported from Karnataka (29.4%) and the least (2.2%) from Andhra Pradesh.

4.1.8.5 Other Diseases

(A) Loose Shell

53.8% of farmers from Karnataka, 45% farmers of Tamil Nadu and Andhra Pradesh farmers reported the problem of loose shell in the shrimp.

(B) Gill Diseases

Gill choking and associated diseases in farmed shrimps were reported by Tamil Nadu (20%) and Andhra Pradesh (2.2%) farmers.

4.1.9 Problems in Shrimp Farming

The results pertaining to the problems associated with resources, socio-economic, technical and farm management are presented in the Table 12.

Table 12. National and State wise Problems in Shrimp Farming

S.No	Problems	India		States (%)					
		Fr	%	AP	TN	GUJ	MAH	KN	KL
(A).	Resources cum Facilities								
1.	Electricity	5	04.1	11.1	00.0	00.0	00.0	00.0	00.0
2.	Water scarcity	6	05	06.6	00.0	00.0	25	00.0	00.0
3.	Lack of lab facility	1	00.8	2.2	20.0	00.0	00.0	00.0	00.0
4.	Seepage	5	04.1	00.0	10.0	00.0	00.0	23.1	00.0
5.	Labour cost	1	00.8	00.0	00.0	00.0	00.0	00.0	6.6
6.	Water quality	10	08.3	00.0	05.0	20.0	50.0	00.0	00.0
(B).	Socio-Economic								
7.	Low market price	26	21.6	26.6	45	26.6	16.6	00.0	00.0
8.	Subsidies	2	01.6	02.2	00.0	00.0	08.3	00.0	00.0
9.	Poaching	2	1.6	00.0	10.0	00.0	00.0	00.0	00.0
(C).	Technical								
10.	Environment problems	2	01.6	04.4	00.0	00.0	00.0	00.0	00.0
11.	Poor technology	2	.6	4.4	00.0	00.0	00.0	00.0	00.0
12.	Water exchange	1	0.8	00.0	00.0	00.0	8.3	00.0	00.0
13.	Survival rate	4	3.3	00.0	10.0	01.6	8.3	00.0	00.0
14.	Algal bloom	4	3.3	00.0	00.0	13.3	16.6	00.0	00.0
(D).	Farm Management								
15.	Seed quality	8	6.6	15.5	00.0	00.0	00.0	00.0	06.6
16.	Loose shell	13	10.8	13.3	45	00.0	00.0	53.8	00.0
17.	Molting	2	02.5	04.4	00.0	00.0	00.0	00.0	00.0
18.	Ammonia	6	05.0	02.2	06.0	00.0	00.0	38.4	00.0
19.	Chemicals	2	01.6	04.4	00.0	00.0	00.0	00.0	00.0
20.	Gill disease	5	04.1	02.2	20.0	00.0	00.0	00.0	00.0
21.	DO	3	02.5	00.0	00.0	00.0	00.0	23.1	00.0

A. Problems of Resources

4.1.9.1 Electricity

11.1% of farmers of Andhra Pradesh reported to have problems due to electricity. None of the farmers from other states reported any such problem.

4.1.9.2 Water Scarcity

6.6% of Andhra Pradesh and 25% of Maharashtra farmers reported availability of water to be a problem in pond culture operations.

4.1.9.3 Lack of Lab Facility

2.2% of the Andhra Pradesh farmers reported the unavailability of lab facilities to test water quality parameters and diseases as a constraint.

4.1.9.4 Seepage

A small percentage of farmers from Tamil Nadu (10%) and Karnataka (23.1%) reported the problems of seepage.

4.1.9.5 Water Quality

A very small number of Indian farmers (8.3%) reported the problem of poor water quality in their ponds. Farmers who reported water quality to be one of the major problems leading to reduction in crop yields accounted for 5% from Tami Nadu 20% from Gujarat, and 50% from Maharashtra.

4.1.9.6 Labour

6.6% of farmers from Kerala reported higher wages for labour affecting the profits. No other state reported similar type of problems.

B. Socio-Economic Problems

4.1.9.7 Low Market Price

21.66% of Indian farmers reported the problem of low market price for the harvested shrimp. 26.6% farmers from Andhra Pradesh, 4.5% of Tamil

Nadu and 26.6% of Gujarat farmers reported the problem fluctuation of market prices for the harvested material.

4.1.9.8 Subsidies

A very low percentage of farmers from Andhra Pradesh (2.2%) and Maharashtra (8.33%) farmers reported the problem of increasing cost of production due to non-availability of subsidy schemes for shrimp farmers.

4.1.9.9 Poaching

From out of the entire country, only the farmers of Tamil Nadu (10%) reported poaching as a problem during shrimp farming.

C. Technical Problems

4.1.9.10 Environmental problems

Fluctuations and in environmental conditions were reported be the reason for less yields by 4.4% of farmers from Andhra Pradesh.

4.1.9.11 Poor Technology

Andhra Pradesh farmers (4.4%) alone reported poor technology to be one of the problems impeding production.

4.1.9.12 Water Exchange

8.3% of farmers from Maharashtra reported that exchange of water was a problem during the farming operations.

4.1.9.13 Survival Rate

10% of farmers from Tamil Nadu, 8.3% from Maharashtra, and 1.66% from Gujarat reported the problems of survival rate of seed, juveniles and adults.

4.1.9.14 Algal Bloom

13.3% of farmers from Gujarat and 16.6% from Maharashtra reported that algal blooms were causing problems in their shrimp ponds.

D. FARM MANAGEMENT PROBLEMS

4.1.9.16 Seed Quality

15.5% of Andhra Pradesh and 6.6% of Kerala farmers reported that the quality of the seed was a factor affecting yield.

4.1.9.17 Molting

4.4% farmers of Andhra Pradesh and 8.6% farmers of Gujarat farmers reported the problems due to improper molting.

4.1.9.19 Ammonia

Presence of Ammonia was reported to be a problem from the ponds of Andhra Pradesh (2.2%), Karnataka (38.4%) and Tamil Nadu (6%).

4.1.9.20 Chemicals

The farmers of Andhra Pradesh (4.4%) reported that the usage of chemicals in shrimp was negatively affecting both yield and quality of material.

4.1.10 Reasons for Crop Loss

The results pertaining to the reasons for the crop loss are presented in Tables 13(a) and 13(b).

23.3% of farmers from India attributed to the seed quality as the reason for crop loss and 19.2% attributed to the poor water quality. Other reasons such as changes in the climate (20.8%) were also responsible for the earlier crop losses. 1.5% farmers attributed to feed quality for the crop losses.

50% of farmers from Indian suffered crop losses due to outbreak of diseases. 56.4% of farmers from west coast region attributed the diseases for

Tab.13 (a). National and Coast-wise Reasons for crop Loss

S.No	Reason	India		East Coast		West Coast	
		Fr	%	Fr	%	Fr	%
1	Seed quality	28	23.3	20	30.7	8	14.5
2	Water quality	23	19.1	16	24.6	7	12.7
3	Climate	25	20.8	18	27.6	1	1.8
4	Poor management	17	14.1	11	16.9	7	12.7
5	Feed quality	2	1.6	1	1.5	6	10.9
6	Poor technology	9	7.5	7	10.7	2	3.6
7	Diseases	51	42.5	29	44.6	31	56.4

* Parenthesis indicates scoring ; Fr- Frequency

Tab.13 (b). State wise Reasons for crop Loss

S.No	Reason	States (%)											
		AP		TN		GUJ		MAH		KN		KL	
		Fr	%	Fr	%	Fr	%	Fr	%	Fr	%	Fr	%
1	Seed quality	17	37.7	3	15.0	1	06.6	6	50	1	07.6	00.0	00.0
2	Water quality	13	28.8	3	15.0	1	06.6	5	41.6	1	07.6	00.0	00.0
3	Climate	17	37.7	1	05.0	1	06.6	5	41.6	1	07.6	00.0	00.0
4	Poor management	9	20	2	10.0	0	00.0	5	41.6	1	07.6	00.0	00.0
5	Feed quality	1	2.2	0	0.00	0	00.0	1	08.3	0	00.0	00.0	00.0
6	Poor technology	5	11.1	2	10	0	00.0	2	16.6	0	00.0	00.0	00.0
7	Diseases	23	51.1	6	30	0	00.0	8	66.6	3	23.0	11	73.3

the crop losses. 73.3% farmers of Kerala, 66.6% farmers of Maharashtra and 51.1% farmers of Andhra Pradesh reported the crop losses due to diseases. All the farmers of Gujarat state were not experienced any disease problem but attributed poor quality of seed, water and unfavorable climate conditions to the earlier crop losses.

4.1.11 Farmers Suggestions for Better Results in Shrimp Farming

The suggestions received from the farmers for yielding better results in shrimp farming are presented in Table 14.

4.1.11.1 Good Aquaculture Practices (GAP)

Majority of Andhra Pradesh (41.6%), Maharashtra (84.2%) and Kerala (88.2%) state farmers had suggested the adoption of Good Aquaculture Practices for yielding better results.

4.1.11.2 Reduction of Cost of Production

The farmers of Tamil Nadu (37.8%) and Maharashtra (15.7%) felt that reduction of cost of production would enable profitable crops.

4.1.11.3 Good Quality Seed

Farmers of Tamil Nadu (32.4%) and Andhra Pradesh (8.3%) farmers suggested that the use of good quality seed would improve the survival rate and reduce the diseases problem.

4.1.11.4 Training Program

Only 4.5% of Karnataka farmers suggested that training programs would facilitate shrimp farmers to improve crop production.

4.1.11.5 Branded Probiotics

The farmers of Gujarat (18.3%) felt that the use of branded probiotics would improve the animal health and thereby reduce the stress related problems.

Table 14. National and Coast-wise Improvements Suggested by Farmers for Better Results in Shrimp Farming

S.No	Suggestion	India (%)	AP (%)	TN (%)	GUJ (%)	MAH (%)	KN (%)	KL (%)
1.	GAP	55.0	33.3	00.0	80	100	61.5	100
2.	Cost of Production	15.8	02.2	31.1	06.6	25.0	00.0	00.0
3.	Good Quality Seed	14.1	06.6	26.6	13.3	00.0	00.0	00.0
4.	Training Programmes	00.8	00.0	00.0	00.0	00.0	07.6	00.0
5.	Branded Probiotics	10.8	04.4	00.0	73.3	00.0	00.0	00.0
6.	Control of shrimp Prices	02.5	00.0	4.4	00.0	00.0	07.6	00.0
7.	Good water Quality	10.8	04.4	00.0	73.3	00.0	00.0	00.0
8.	Technical Support	5.8	02.2	00.0	00.0	00.0	46.1	00.0
9.	Disease Diagnosis	11.6	00.0	20	20.0	00.0	00.0	13.3
10.	Others	31.6	26.6	00.0	100	00.0	46.1	00.0

*Others: Lab facility, subsidies, and integrated farming ;GAP-Good Aquaculture Practices

4.1.11.6 Control of Shrimp Prices

5.4% of farmers from Tamil Nadu and 4.5% of farmers from Karnataka felt that control measures over the fluctuations of shrimp prices would yield better results.

4.1.11.7 Good Water Quality

18.3% of farmers of Gujarat and 5.5% of farmers of Andhra Pradesh had suggested that maintenance of good water quality in shrimp culture ponds would provide a better yield.

4.1.11.8 Technical Support

Farmers of Karnataka (27.2%) suggested that necessary technical support in improving the culture practices scientifically would yield positive results.

4.1.11.9 Disease Diagnosis

The farmers of Tamil Nadu (24.3%), Gujarat (5%) and Kerala (11.7%) had suggested that proper diagnosis of causative organisms and followed therapeutic treatment would enable the farmers to obtain better yield.

4.1.11.10 Other suggestions

Farmers from Andhra Pradesh (33.3%), Gujarat (33.2%) and Karnataka (27.2%) suggested the provision of subsidies to purchase materials for shrimp culture operations, availability of lab facilities and integrating farming with poultry would improve the shrimp production with better results.

4.1.12 Relationship between the usage of Antibiotics, Probiotics, Chemicals, Stocking Density, Culture Condition, Source of Guidance, Training Experiences, Awareness of Banned Antibiotics and Awareness of MPEDA Guidelines

In order to measure the relation between usage of antibiotics, probiotics, chemicals, stocking density, culture condition, source of guidance, training experience, awareness of banned antibiotic and awareness of

MPEDA guidelines, significance tests such as 'Somers'd and Kendall's tau-c and tau-b were used to test only responded (valid) farmers. tau-c test was used to when number of categories of row and column variables are not equal and tau-b for equal number of categories and the results are presented in Table.15

Application of antibiotics had non-significant relationship with the stocking density, culture condition, source of guidance, training experience, awareness of banned antibiotics and awareness of MPEDA guidelines.

Application of molt inducers and probiotics had significant relationship with stocking density at 3% and 4% levels respectively. Molt inducers had 1% level of significant relationship with training experience of the farmer.

The usage of Probiotics had significant relationship at 1% level with the culture conditions prevailing at the shrimp farms.

4.1.13 Relationship between Education, Occupation, Farm Area, Water Source, Usage of Antibiotics and Chemicals

In order to measure the relationship between the application of antibiotics, chemicals, stocking density, culture conditions, source of guidance, training experience, awareness of banned antibiotics and awareness of MPEDA guidelines, tests of significance such as Somers'd and Kendall's tau-c' were used for only responded (valid ones) farmers. The results are presented in Table 16.

The variables such as Occupation of the farmer and farm area did not possess significant relationship with the use of antibiotics and chemicals whereas education level of the farmer had 1.7% level of significant relationship with the use of probiotics. The source of water had 0.7% level of significant relationship with the use of probiotics and molt inducer and the correlation coefficient values were 0.491 and 0.195 respectively.

Table 15. Correlation between Antibiotics, Chemicals and other Variables

S.No.		Antibiotics			Molt inducers			Sanitizers			Probiotics		
		NR	No	Yes	NR	No	Yes	NR	No	Yes	NR	No	Yes
1.	Stocking Density	NS			S:0.003 V:0.245			NS			S:0.004 V:(-0.196)		
	a) > 15	4	5	2	6	-	5	5	-	6	2	-	9
	b) 13 – 15	12	16	-	10	7	11	7	2	19	-	1	27
	c) 8 – 12	30	9	-	20	1	18	22	-	17	6	4	29
	d) 5 – 7	34	7	1	21	-	21	27	-	15	9	8	25
2.	Culture Condition	NS			NS			NS			S:0.001 V:0.289		
	a) Poor (or) below Optimum	47	17	1	32	3	30	34	-	31	9	12	44
	b) Optimum	33	20	2	25	5	25	27	2	27	8	1	46
3.	Source of guidance	NS			NS			NS			NS		
	a) Self (or) Fellow Farmers	25	6	1	12	-	20	12	-	20	3	3	26
	b) Technician	45	29	-	40	6	26	41	2	31	10	10	54
	c) Professional	10	2	2	5	-	9	8	-	6	4	-	10
4.	Training experience	NS			S:0.001 V:0.341			NS			NS		
	a) No	51	33	1	50	8	27	53	2	30	16	10	59
	b) Yes	29	4	2	7	-	28	8	-	27	1	3	31
5.	Awareness of ban	NS			NS			NS			NS		
	a) No	10	3	1	10	-	4	10	-	4	5	3	6
	b) Yes	70	34	2	47	8	51	51	2	53	12	13	84
6.	Awareness of MPEDA guidelines	NS			NS			NS			NS		
	a) No	46	10	2	27	4	27	29	-	29	15	6	37
	b) Yes	34	27	1	30	4	28	32	2	28	2	7	53

NS – Non Significant; S – Significant; V – Correlation Coefficient Value

Table.16 Correlation between usage of antibiotics, chemicals and other variables

S.No	Variables	Probiotics			Molt inducers			Sanitizers			Antibiotics		
		NR (0)	NO (1)	YES (2)	NR (0)	NO (1)	YES (2)	NR (0)	NO (1)	YES (2)	NR (0)	NO (1)	YES (2)
1.	Education	S :0.017 V: 0.198			NS			NS			NS		
	Primary (1)	2	8	16	14	1	11	16	-	10	19	7	-
	Secondary (2)	11	3	34	25	3	20	21	2	25	35	13	-
	Graduates (3)	4	1	35	17	4	19	23	-	17	23	14	3
	Professional (4)	-	1	5	1	-	5	1	-	5	3	3	-
2.	Occupation	NS			NS			NS			NS		
	Aqua+Agri (1)	7	1	18	14	-	12	15	1	10	22	3	1
	Aqua Only (2)	7	12	50	38	5	26	40	1	28	44	24	1
	Aqua+Consult (3)	3	-	22	5	3	17	6	-	19	14	10	1
3.	Farm Area	NS			NS			NS			NS		
	0 – 2 (1)	9	8	27	27	3	19	31	1	12	29	13	2
	3 – 5 (2)	3	3	35	18	1	22	17	1	23	28	13	0
	> 5 (3)	5	2	17	12	4	8	13	-	11	12	11	1
4.	Water Source	S : 0.00 V: 0.491			S : 0.007 V: (-0.195)			NS			NS		
	Bore well (1)	3	11	17	18	-	13	21	-	10	24	5	2
	Creek (2)	14	2	73	39	8	42	40	2	47	56	32	1

Significance Test for only those Responded Farmers (valid cases)

S-Significance, NS-Non-Significance, V-Correlation coefficient value

4.1.14 Relationship between Profile of Characteristics of Shrimp Farmer and Shrimp Farms, Intensity of Shrimp Farming, Source of Guidance, Stocking Density, Culture Conditions, Usage of Antibiotics and Chemicals

In order to measure the relationship that exists between the profile of shrimp farmers and shrimp farms, intensity of shrimp farming, source of guidance, usage of antibiotics and chemicals, the correlation coefficients have been worked out and tested for its statistical significance that are presented in Table 17.

Education, occupation, soil type, farm experience, farm area, water spread area, source of guidance, stocking density and culture conditions did not possess significant relationship with the application of antibiotics, whereas water source and farm type had negative correlation with the usage of antibiotics at 5% level of significance and the correlation coefficient value - 0.368 for each.

The source of water and usage of antibiotics had 5% level of significant relationship with correlation coefficient of – 0.368. The source of water and usage of probiotics had 1% level of significant relationship with correlation coefficient of 0.491. The education level, soil type, farm area, stocking density and culture conditions and usage of probiotics had 1% level of significance.

The usage of chemicals such as molt inducers and sanitizers did not reveal any significant relationship with the profile of characteristics of shrimp farmers and shrimp farms except stocking density of the farms that possess significant relationship at 1% level with correlation coefficient of 0.334.

4.1.15 Relationship between Usage of Antibiotics and Chemicals

In order to measure the relationship between usages of antibiotics, probiotics and molt inducers, tests of significance were used and the results are presented in Table 18. The results showed that none of them were significantly related.

Table 17. Correlation between Variables and Usage of Antibiotics and Chemicals

	Stocking Density	Culture Condition Average score	Antibiotics	Molt inducers	Sanitizers	Probiotics
Education	NS	S V: 0.204	NS	NS	NS	S V: 0.273
Occupation	NS	NS	NS	NS	NS	NS
Soil type	NS	NS	NS	NS	NS	S V: (- 0.266)
Water source	S V:(-0.286)	S V: 0.428	S V:(- 0.368)	Ns	Ns	S V: 0.491
Experience	NS	NS	NS	NS	NS	NS
Farm Area	NS	S V: 0.199*	NS	NS	NS	NS
Water spread area	NS	NS	NS	NS	NS	S V: 0.257
Farm type	NS	NS	S V:- 0.368*	NS	NS	NS
Source of guidance	NS	NS	NS	NS	NS	NS
Stock density	NS	S V:(- 0.280)	NS	S V: 0.334	NS	S V:(- 0.267)
Culture condition	NS	NS	NS	NS	NS	S V: 0.289

S - Significance at 0.05 (5%) level.

V – Correlation Coefficient value

NS – Non Significance at 0.01 (1%) level

Tab 18. Correlation between Usage of Antibiotics and Chemicals

	Antibiotics				Molt Induces		
	NR (o)	No (1)	Yes (2)		NR (o)	No (1)	Yes (2)
Probiotics							
NR (0)	15	1	1	17	-	-	-
No (1)	11	2	34	47	0	0	8
Yes (2)	54	-	2	56	-	2	53
Antibiotics							
NR (0)	-	-	-	-	39	1	40
No (1)	-	-	-	-	17	7	13
Yes (2)	-	-	-	-	1	-	2

* None of them are significantly related with one another

NR-No Response

4.2 Estimation of Oxytetracycline Withdrawal Period in *Penaeus monodon*

Results of the present study on tissue residues of OTC are presented in Table 19 and Figs. 4-6. It has been observed that the amount of OTC on day 1 after cessation of medicated feed was 4.842 ppm, which reduced gradually to reach the lowest of 0.001ppm on day 15. Tissue residues were totally abolished by the 16th day. ANOVA indicated that there was no significant variation between the control and treatment means of the observed F value with respect to the length-weight relationship (Table 20).

4.3 Hazard Analysis of Shrimp Feed Samples

The microbiological analysis of all the commercial shrimp feeds revealed that most of the feeds were contaminated with aerobic bacteria. The total aerobic plate count of the feeds ranged from 10^2 - 10^4 cfu/g (Table 21). The feed samples tested for coliform contamination (Plate 13) showed the contamination of faecal coliform ranged from 10^2 - 10^4 C.F.U./g. Amongst the total two commercial feeds (Feed B and Feed J) were not contaminated with any faecal coliform. All the commercial shrimp feeds except one (Feed C) were not contaminated with *Staphylococcus aureus*.

The feed sample - C (Table 21) showed maximum contamination with coliform, and was further tested for the *E. coli* contamination cultured in EMB agar. The results confirm the presence of *E. coli* (Plate 14). The biochemical tests on indole production of confirmed the presence of the bacteria.

The results of the physical hazard analysis (Table 22) revealed that majority of the feed samples were not contaminated with any of the physical hazards such as leaves, plastic, stick, etc. But, three feed samples possessed a poor texture, being more powdery as compared to other feeds that had a good pellet structure with proper binding. One commercial feed (Feed-D) revealed to possess an excellent texture, not having any of the physical contaminants.

Table 19. Concentration of Oxytetracycline (OTC) detected from shrimp muscle tissues using HPLC

S.No	Experiment	OTC Concentration During Post-Medication Period (Day)				
		1st	7 th	14 th	15 th	16 th *
1	Control	0.000	0.000	0.000	0.000	0.000
2	Treatment	4.842	2.563	0.012	0.001	0.000

* 16th Day – 24th October 2004

Table 20. ANOVA between Length and Weight of Shrimps treated for estimation of Oxytetracycline Withdrawal Period

**Anova: Weight
SUMMARY**

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
W=T1	25	303.6	12.144	2.40367
W=T2	25	323.14	12.9256	2.69055
W=C	25	305.59	12.2236	2.75165

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	9.25032	2	4.62516	1.76851	0.17792	3.12390114
Within Groups	188.301	72	2.61529			
Total	197.551	74				

**Anova: Length
SUMMARY**

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
L=T1	25	296.74	11.8696	0.24357
L=T2	25	305.78	12.2312	0.83294
L=C	25	301.38	12.0552	0.68634

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	1.63482	2	0.81741	1.39106	0.25542	3.12390114
Within Groups	42.3084	72	0.58762			
Total	43.9432	74				

* No significant difference between Control and Treatment means

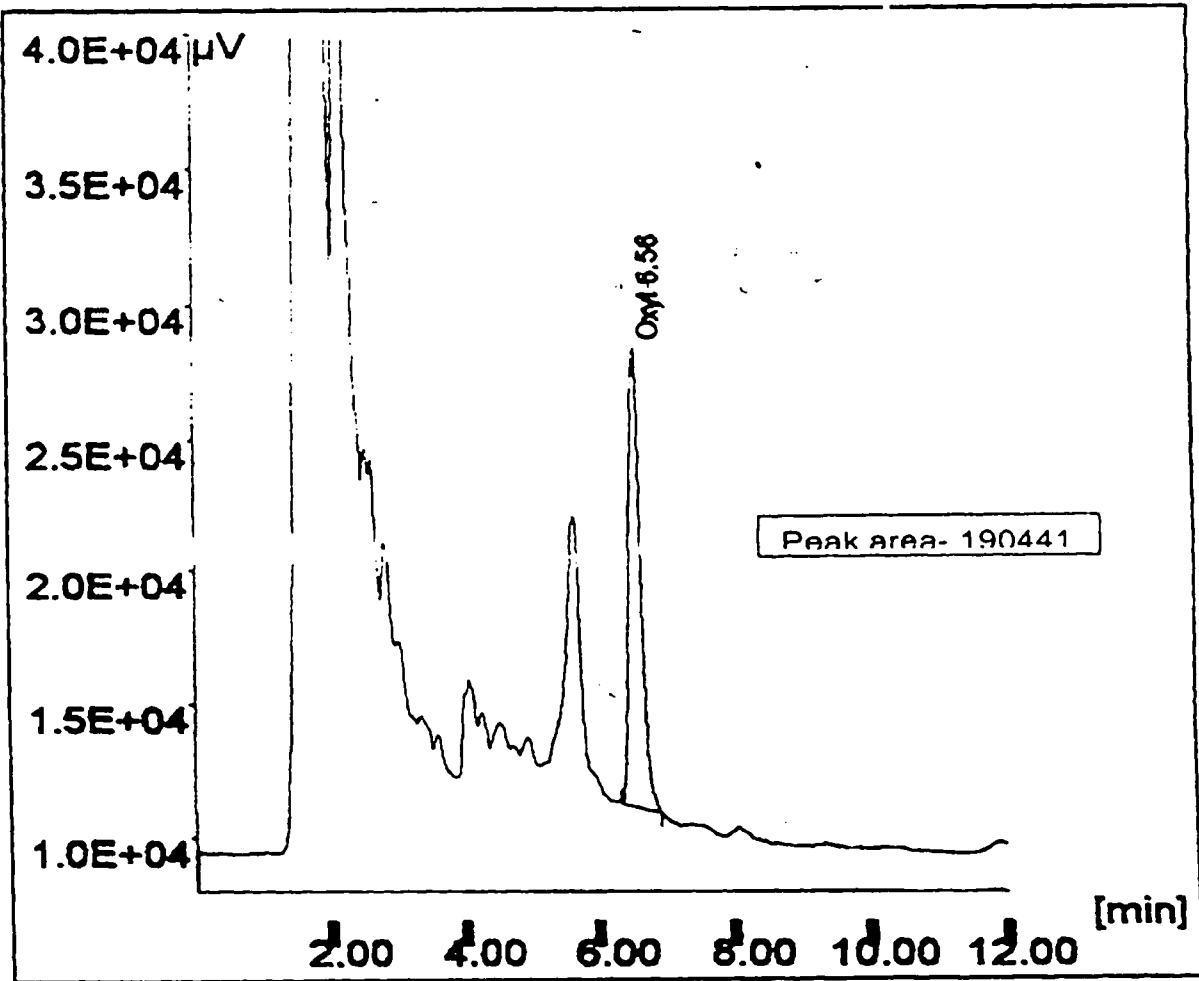


Fig. 4. Chromatogram of residual oxytetracycline in the muscle tissue of treated shrimps on 1st day after cessation medicated feed

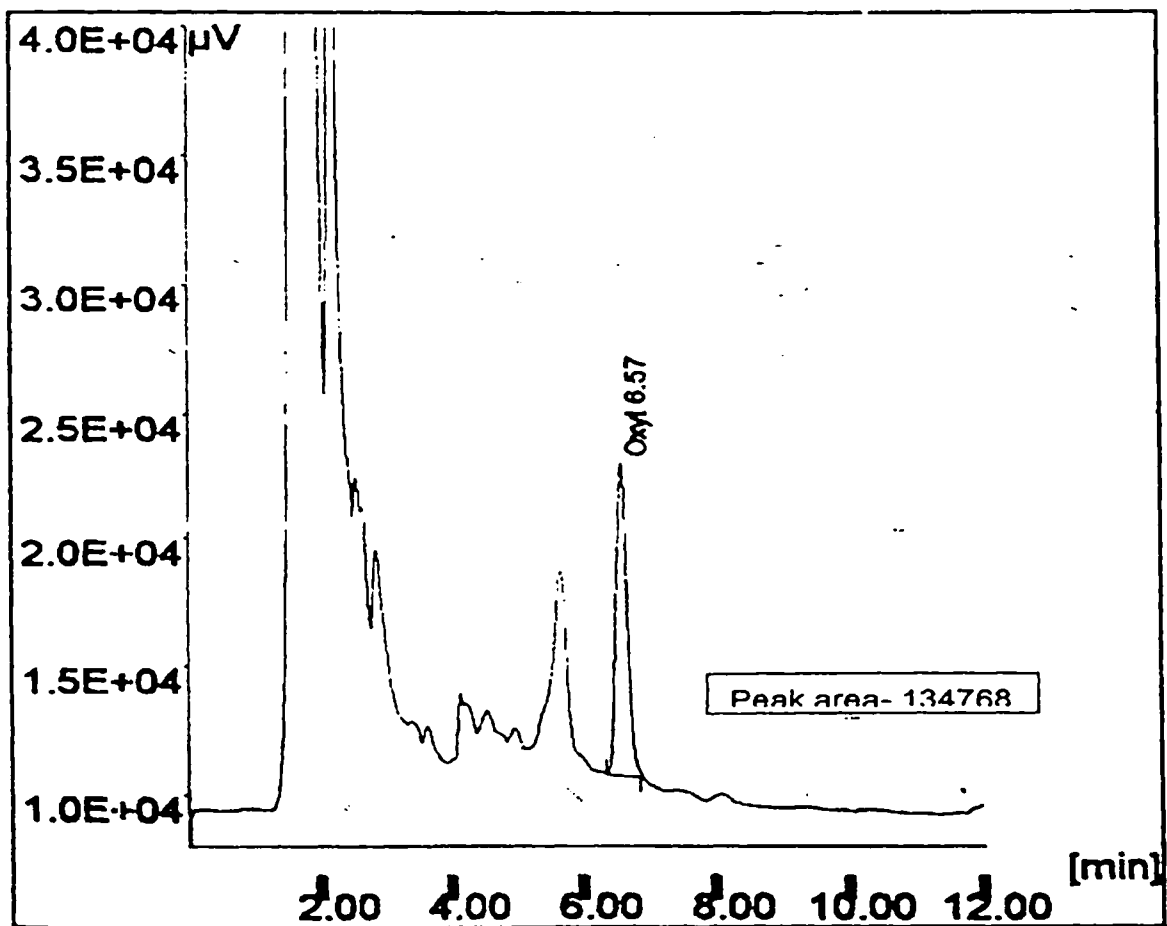


Fig. 5. Chromatogram of residual oxytetracycline in the muscle tissue of treated shrimps on 7th day after cessation medicated feed

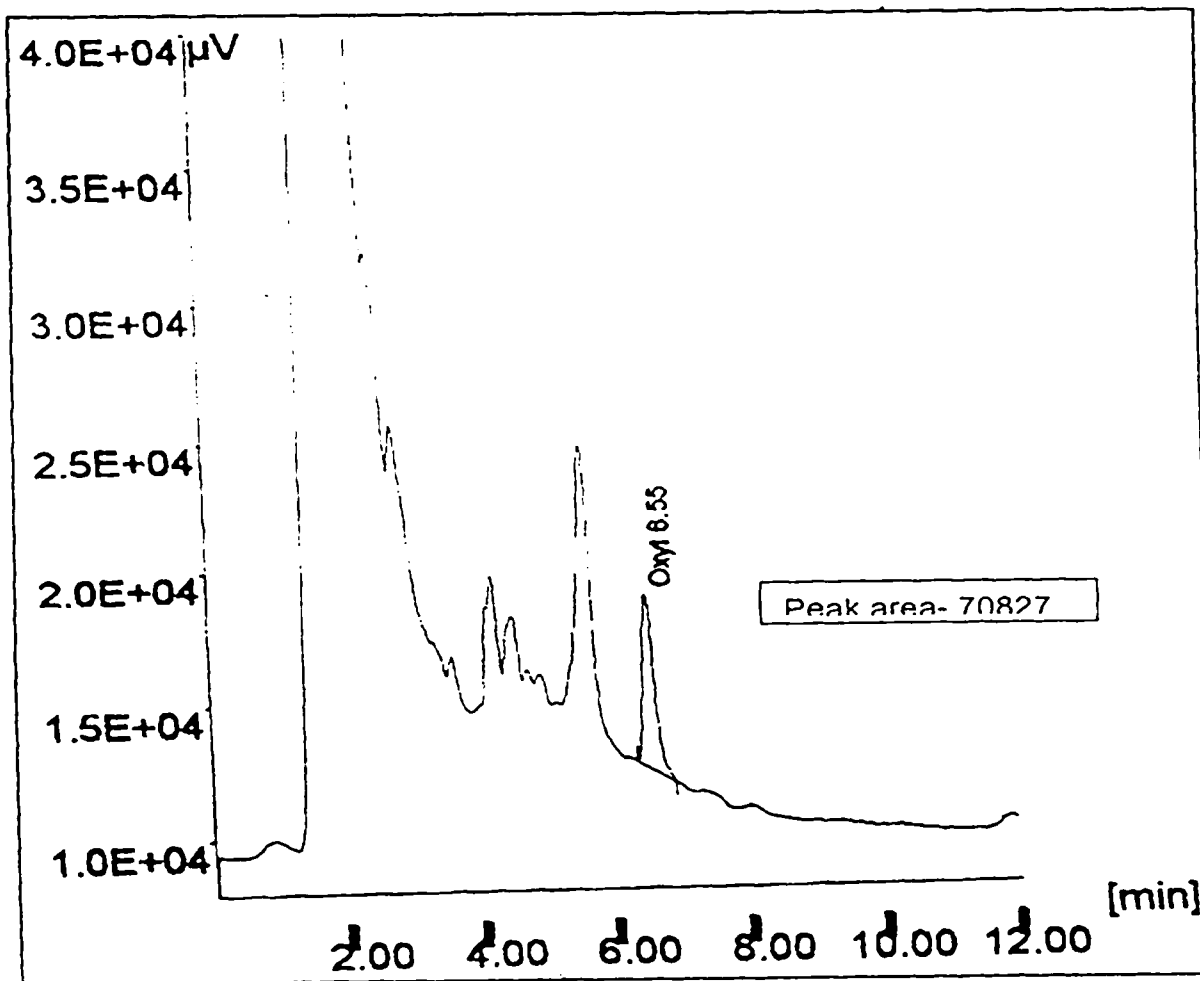


Fig. 6. Chromatogram of residual oxytetracycline in the muscle tissue of treated shrimps on 15th day after cessation medicated feed

Table 21. Microbiological Hazard Analysis of Commercial Shrimp Feeds (n=30)

S.No	Feed Sample (N= 30)	Feed Specification	Total Aerobic Plate Count (cfu/g)	Total Coliform count (cfu/g)	<i>Staphylococcus aureus</i> (cfu/g)
1	A	Grower	1.2X10 ⁴	1.0x10 ⁴	NIL
2	B	Grower	1 X 10 ²	NIL	NIL
3	C	Grower	8 X 10 ²	4.2x10 ²	1x10 ²
4	D	Grower	1.2 X 10 ³	NIL	NIL
5	E	Grower	1.1 X 10 ²	1.0x10 ²	NIL
6	F	Grower	2.1 X 10 ⁴	1.4x10 ⁴	NIL
7	G	Grower	1.8 X 10 ²	1.0x10 ²	NIL
8	H	Grower	3.2 X 10 ³	2.1x10 ³	NIL
9	I	Grower	2.4 X 10 ³	1.3x10 ³	NIL
10	J	Grower	1.0 X 10 ³	NIL	NIL

n – total number of samples

Table 22. Physical Hazards of Commercial Shrimp Feeds

Sl. NO	Feed (n=30)	Texture (Binder)	Wood	Stones	Metal	Sand	Plastic	Glass	Leaves	Sticks
1	A	Good	-	-	-	-	-	-	-	+
2	B	Good	+	-	-	+	-	-	+	+
3	C	Poor	-	+	-	+	-	-	+	+
4	D	Excellent	-	-	-	-	-	-	-	-
5	E	Poor	-	-	-	+	-	-	-	-
6	F	Good	-	-	-	-	-	-	-	-
7	G	Good	-	-	-	-	-	-	-	-
8	H	Poor	-	-	-	+	-	-	+	-
9	I	Good	-	-	-	+	-	-	-	+
10	J	Good	-	+	-	-	-	-	+	-

Scale : Poor -1, Good - 2, Excellent - 3.

Symbol '-' Indicates Absent, '+' Indicates Present

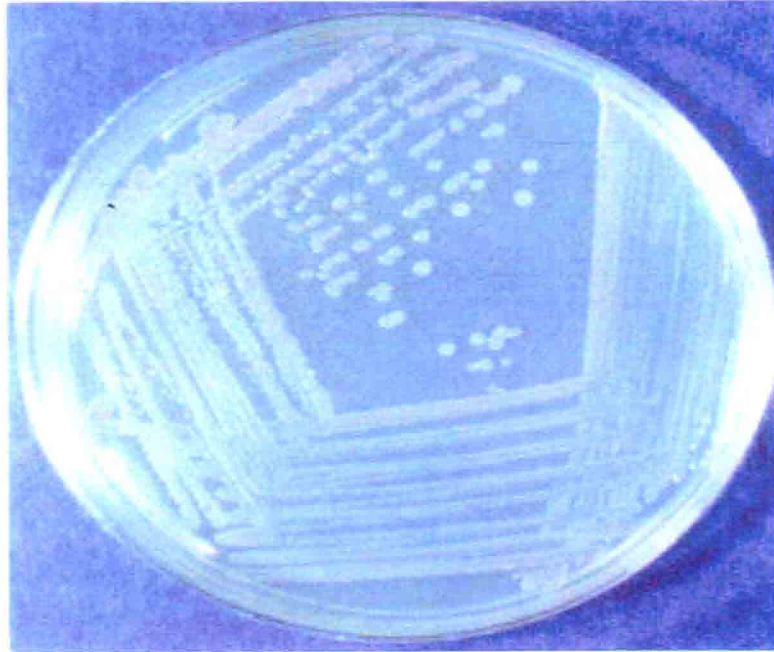


Plate 13: Showing coliforms isolated from shrimp feed



Plate 14: Showing *E. coli* isolated from shrimp feed

4.4 DEVELOPMENT OF A HACCP FRAMEWORK FOR INDIAN SHRIMP FARMING TO PREVENT ANTIBIOTICS RESIDUES

4.4.1 Assembling of HACCP team

HACCP team consists of personnel having working knowledge of shrimp farming. The field survey indicated that majority (61.7%) of shrimp farms in India were depending on feed technician for the technical help for the on-going shrimp farming operations and a few were depending on the professionals of Government organizations. Most of the shrimp farmers (40%) were having the education back ground of matriculation or higher secondary and possess the shrimp farming experience (65.8%) of more than three years. For construction the HACCP team, feed technician, the farm owner or farm manager who is working agent or company representative who is purchasing the harvested shrimp as where most of the Indian shrimp farmers were selling the harvested shrimp either directly (58.3%) or indirectly through agents (41.7%). As far as possible and the availability, the company people to be incorporated in the HACCP team. In addition to the three members, two more personnel having experience of HACCP principles and shrimp health management practices especially having complete knowledge on disease diagnosis use of antibiotics and other pharmacologically active substances. As far as possible it is essential to include the expert from the Government organizations for yielding better results.

Based on the filed survey findings the following composition was identified to be essential assembled for HACCP team at the farm level.

1. The farmer (or) Farm manager
2. Qualified/Certified feed technician
3. Representatives of processing plant/qualified or certified marketing agent
4. Qualified/certified shrimp health specialist
5. HACCP expert for shrimp farming

4.4.2 Describing the Produce / Product

The survey findings indicated that all of the Indian shrimp farmers except from W.B. & Orissa were stocking hatchery seed for their shrimp operations. Most (85.8%) the shrimp farmers in India were using commercial shrimp feed and creek water (73.3%) for the shrimp culture. Majority (90.8%) of the shrimp farmers in India were selling their product as Head-on type. Antibiotics and chemicals would be applied during grow-out operations. Harvesting would be operated by nets and the harvested raw material would be iced for chilling prior to marketing either directly or through middlemen.

4.4.3 Identification of End Users

The market reports showed that most of the shrimp cultured product was exported to USA and Japan and European Union (Anon, 2005) and the end-users were identified as consumers in European Union, USA and Japan. The international end user is identified as general public.

4.4.4 Construction of flow diagram

A flow diagram describing the major steps in shrimp farming operations of India was constructed and presented in Fig 2 (a) and Fig.2(b).

4.4.5. On site verification of the flow diagram

All the stages mentioned in the flow diagram was verified and the survey results revealed that the flow diagram covered all the basic aspects of shrimp farm operations. The results of all the east coast and west coast regions shrimp farms were indicated that the shrimp farm operations (Fig.2 (a) and Fig.2 (b)) were in similar pattern in most of the shrimp farm.

4.4.5.1 Site selection

Even though survey was conducted in the existing shrimp farms the site selection was included as first step, despite the fact that it would be useful only in constructing the new shrimp pond. Majority (43.3%) of the Indian shrimp farms were sandy loamy type soils and sandy clay soils (39.2%).

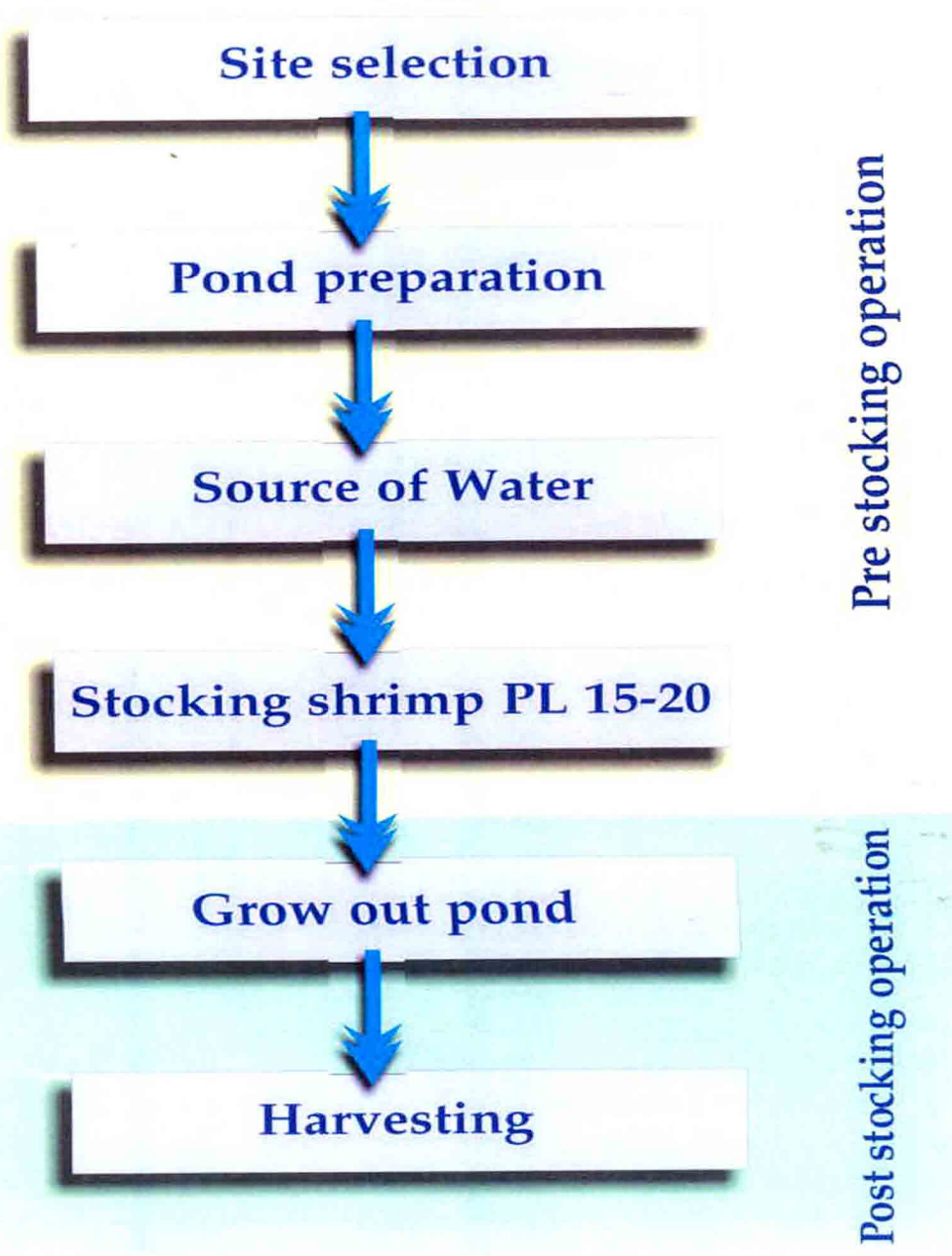


Fig. 7 (a). Flow chart showing shrimp farming operations in India

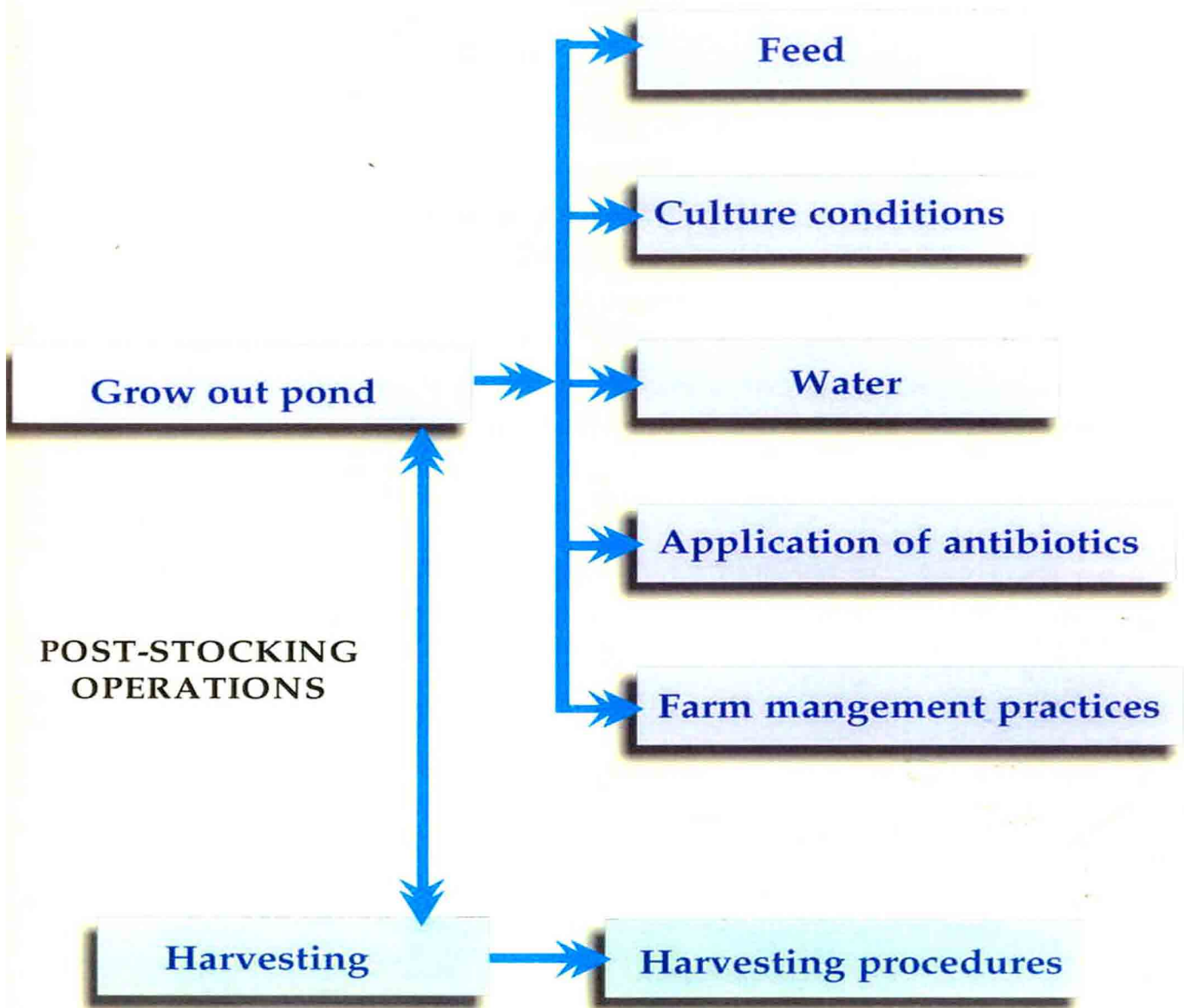


Fig 7 (b). Flow chart showing post-stocking operations at shrimp grow-out farms

Majority of the farmers possess 0-2 ha of farm area, albeit without much selectivity.

4.4.5.2 Pond preparation

Almost all the shrimp farmers in India were using the fertilization of N, P, and K for their pond preparations. The results indicated that 76.7% of shrimp farmers in India were practicing two crops per year with an average of 120-150 days of culture period. The pond preparation was mainly carried out during the interval of crops and during the starting stages and before stocking the post larvae in the pond. The main sources for inorganic fertilizers were fertilizer dealers. Organic manuring was also carried out depending on the availability of cattle or cow dung manure.

4.4.5.3 Water Source

Majority (73.3%) of the Indian shrimp farmers were using the creek water as the major water source. The farmers in Andhra Pradesh (60%) were using the ground water especially in the districts of East Godavari and West Godavari and some parts of Nellore.

4.4.5.4 Stocking of shrimp post larvae

All the shrimp farmers in the present study areas were using the hatchery seed as main source in the shrimp farms. Majority (88.3%) of the shrimp farmers were stocking post larvae of 15-20 days of seed.

4.4.5.5 Feed

Majority (88.3%) of the shrimp farmers were using the commercial feeds of different branded feeds. The results showed that around half of the farmers (57.5%) were obtaining Feed conversion ratios (FCR) below 1.5.

4.4.5.6 Water

The survey findings showed that majority (54.2%) of the shrimp farmers were maintains poor culture conditions. The water quality parameters like pH, salinity, turbidity, dissolved oxygen, plankton and water colour were

measured by the most of the farmers (54.2%) for every monthly. Majority of the shrimp farmers (91.7%) were maintaining the optimal depth.

4.4.5.7 Application of Antibiotics

Several types of antibiotics were used indiscriminately for therapeutic purpose only. The present survey results indicated that 30.8% of the farmers were not using the any type of antibiotics and majority (66.7%) of Indian shrimp farmers selected for the present study refrained from revealing any information regarding the application of antibiotics. Only negligible (2.5%) number of farmers revealed the fact of application of antibiotics.

4.4.5.8 Farm Management Practices

The management practices include handling of shrimp, water exchange, usage of aerators and other practices which were not covered in this grow out operations mentions in the other four stages mentioned earlier. The management practices would reflect the standard in line with the government guidelines. The survey findings showed that the farmers were using molt inducers (45.8%) and sanitizers (47.5%). Majority (75%) of the Indian shrimp farmers were using them to maintain health conditions of the shrimps. Most of the farmers were testing the seed prior stocking in the shrimp ponds. Majority of the seed was tested by PCR for white spot syndrome virus, formalin test for testing the test.

4.4.5.9 Harvesting

The harvesting of shrimps (57.5%) would be carried out after 120 days of culture period. The harvested shrimp would be sold to either company directly by the farmer or to the agents.

4.4.6. Hazard analysis

The identified hazard is banned Antibiotic residues were analyzed for presence of residues of the banned antibiotics from the samples of harvested shrimp at each step in shrimp farming operations. The banned antibiotics like

Chloramphenicol and Nitrofurantoin and its metabolites were of major concern by the importing countries because of their concern to human health.

The field study carried out in several shrimp farms in India had identified the factors and situations leading to the application of antibiotics. Most of the farmers were using molt inducers, sanitizers and probiotics as a part of management practices. There was significant relationship (2.45% and 1.95% level) of application of molt inducers and probiotics with that of stocking density.

The antibiotic hazard might enter the grow out operations at the time of pond preparation. The occurrence of this hazard would depend on the farm management practices, seed testing, and pond culture and soil type. The mixing of antibiotics at the on-farm feeding was the major source of this hazard. Even though majority (88.3%) of Indian farmers knew about the banned antibiotics, the ignorance of few farmers (17.8% in AP) about awareness of banned antibiotics is leading to the occurrence of this antibiotic hazard. Microbial and physical hazards present in the shrimp feed would be influencing the application of antibiotics.

4.4.7 Identification of Critical Control Point (CCP's)

Based on the survey findings and with aid of CCP decision tree (Table.23), the following phases/steps in shrimp farming were identified as critical control points.

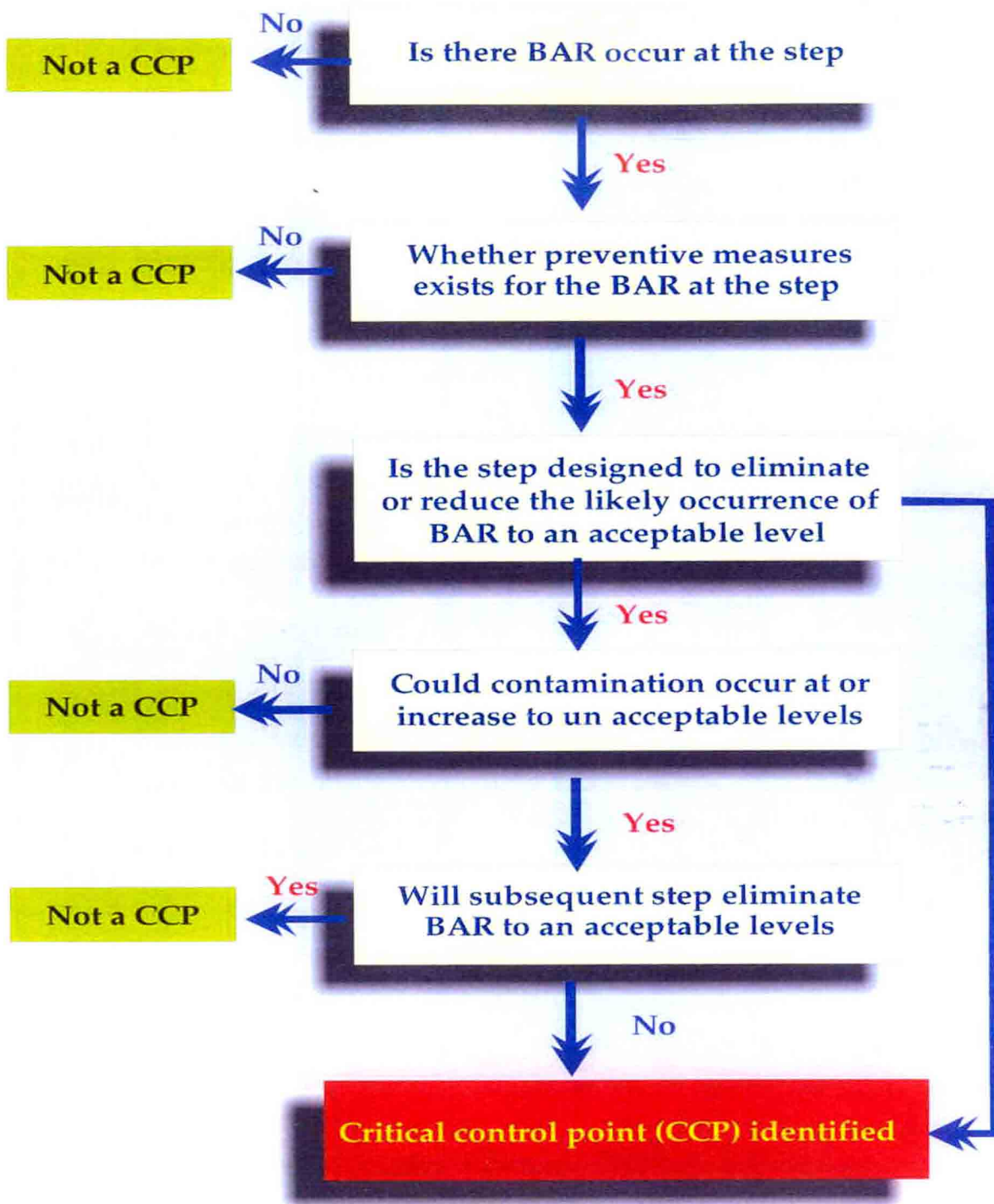
1. Site selection
2. Pond preparation
3. Water source
4. Shrimp seed
5. Shrimp feed
6. Water quality
7. Application of antibiotics
8. Farm management practices
9. Harvesting

Table 23. Analysis of Different Steps for Critical Control Points (CCP's) Identification in Shrimp Farming Operations

S. No	Steps in Shrimp Farming	Significance	Justification	Preventive Measures	CCP
1	Site selection	Yes	Shrimp site may be polluted the neighbouring farms	Proper site selection by testing the soil samples for BAR	Yes
2	Pond preparation	Yes	Usage or presence of BAR will present in the previous crop. Application of chemicals during pond preparation	Sun dry the pond for appropriate period and follow the guidelines of MPEDA for pond preparation	Yes
3	Water source	Yes	The source of water may be contaminated with BAR	Use pre tested water and maintain reservoir tanks	Yes
4	Shrimp seed	Yes	The hatchery produced shrimp might have residues of BAR. Post larvae also infected with viral particles which in turn pose problem of BAR	Testing of post larvae for disease and BAR	Yes
5	Feed	Yes	The feeds used of shrimp forms would be mixed with banned Antibiotics	Don't use banned antibiotics. Use feed probiotics of good quality	Yes
6	Water Quality	Yes	The water quality will in turn responsible for the application of banned antibiotics	Don't use banned antibiotics and chemicals	Yes

7	Application Antibiotics	Yes	Indiscriminate use of pharmacologically active substances and chemicals will pose BAR	Don't use banned Antibiotics, drugs & chemicals for prophylactic purposes	Yes
8	Farm Management Practices	Yes	The health management practices, feed management, water quality management, standard operations includes handling of shrimp will lead to applications banned antibiotics	Use good aquaculture practiced for suitable development. Don't use banned antibiotics	Yes
9	Harvesting	Yes	During the last days of crop harvesting, chances of using foreign feed substances like snails, oyster etc, which may have BAR	Don't use foreign feed substances without prior testing. Maintain probable with drawl time by postponing the harvesting crop	Yes

BAR: Banned Antibiotic Residues



*BAR-banned antibiotic residues

Fig 8. The critical control point (CCP) decision tree

4.4.7.1 Site Selection

The selection of site for shrimp farming was identified as a critical control point as proper site selection would help in selecting good soil for shrimp pond. Good pond soil will enhance good pond bottom and optimal culture conditions and indirectly helps in preventing the usage of application of antibiotics. There is also the possibility of contamination by leakage/ discharge from agricultural farms etc. Proper site selection will also helps in selecting proper site and this critical control point could be considered only for construction of new shrimp farms and would not applicable to existing farms.

4.4.7.2 Pond Preparation

There is more probability of presence of traces of antibiotic residues and other pharmacologically active substances in the soil and likely to enter the system. During the Pond preparation for the next crop, it is essential to take necessary preventive measures at this step.

4.4.7.3 Water Source

As, most of the shrimp farmers in India were using the creek water or surface water, there is more possibility of occurrence of banned antibiotic residues contamination from the water source.

4.4.7.4 Shrimp Seed

The seed used for stocking in shrimp ponds is the main source diseases such as white Spot Syndrome Virus (WSSV). Even though, majority of the shrimp farmers all over India were conducting PCR tests on post larvae for white spot disease, the field survey results showed that most of the farmers were reporting the problems of white spot and other diseases. Because of this farmers were compelled to use antibiotics and other pharmacologically active substances indiscriminately even though none of the antibiotics will do nothing with the viral diseases. There would be more possibilities of contamination of banned antibiotic residues from hatchery.

4.4.7.5 Feed

The feeding strategies employed at farm level would have possibilities of occurrence of antibiotics. The wastage of feed would lead the pollution water quality and lead to possibilities of occurrence of diseases. As most of the antibiotics were applied at farm level by mixing with the feed on daily basis.

4.4.7.6 Water Quality

The conditions exists in the water would have more possibilities of applying the chemicals and drugs to maintain optimum water quality parameters. The unlabeled drugs and chemicals would lead to the possibilities of occurrence of banned antibiotic residues.

4.4.7.7 Application of Antibiotics

Indiscriminate use of antibiotics and other pharmacologically active substances as routine practice would pose more probability of occurrence of banned antibiotic residues.

4.4.7.8 Farm Management Practices

The management practices that include handling of shrimps, maintenance of storage conditions for feeds, standard operation procedures to maintain good hygienic and sanitation and health management practices will have more probability to use antibiotics in the shrimp farms.

4.4.7.9 Harvesting

It is the step where quality of the shrimp would be contaminated with the microbial hazards. The methods used for harvesting shrimps from the ponds would affect the quality of the shrimps. The marketing of the shrimps would also indirectly affect the quality of the harvested shrimp

4.4.8 Establishment of Critical Limits for each CCP

For establishing critical limits for the each identified CCP, the guidelines and standards fixed by the Marine Products Export Development Authority (MPEDA), India were taken as basis. The results of the survey, antibiotic withdrawal time and analysis of shrimp feed were used to fix the critical limits for each CCP.

4.4.8.1 Site selection

The soil testing for banned antibiotic residue to be done prior to site selection and the soil should not contain any left over traces of banned antibiotic residues. It is appropriate to select the site for shrimp farming free from any contamination from neighboring farms. The soil construction to be followed as per the guideline issued by the good farm construction. The ideal soil texture for construction and water retention are clay loam or loam. The survey indicated that majority of the shrimp farmers of India contain sandy loamy. The management practices that to be followed should not include any banned antibiotics.

4.4.8.2 Pond Preparation

The pond preparation practices generally include use of N, P, K fertilizers and manuring of cow or cattle dung for the developing desired level of plankton and algal bloom which in turn useful as source of natural feed for shrimp seed as for the. It is essential to check that both organic and inorganic fertilizers should not contain any traces of banned antibiotics. It is essential to use only labeled fertilizers. Prohibit the use of unlabeled products of any kind.

4.4.8.3 Water Source

The water used for stocking the shrimp pond should not contain any residues of the banned antibiotics. Majority of the shrimp farmers indicated that they were using creek water and a few farmers in Andhra Pradesh especially Godavari district were using the ground water. Proper checking of the water for contamination for disease causing pathogen such as viral and

bacteria which may lead to the application of chemicals and antibiotics. It is essential that water used for the shrimp culture should not contain any pathogens.

4.4.8.4 Shrimp Seed

The post larvae to be stocked in the shrimp culture have to be tested compulsory with PCR for white spot syndrome virus (WSSV). For the PCR results to be reliable only authorized and certified labs should be consulted. As most of the farmers in the survey indicated that they are doing PCR testing the seed and the problems were cumulative. It indicated the reliability of the results tested by the PCR labs. The seed also should accompany with certificate from the hatchery stating the non-usage of banned antibiotics during for production of post larvae.

4.4.8.5 Shrimp Feed

Majority of the shrimp farmers were using commercial feeds for their grow-out operations. The results of feed analysis revealed that majority of the feeds did not possess the proper texture and had loose binding capacity. It is essential to use the feeds with good structure and binding capacity that should make the pellets in compact for about 2 to 3 hours in the water column. The results of the feed analysis for microbiological hazards indicated that there is a possibility of break down of sanitation and hygiene. The feed should be handled in proper hygienic conditions. It should be kept in good storage conditions. The increase of temperature and moisture would have more possibility of contamination of pathogenic bacteria and also development of mycotoxins especially by *Aspergillus*. Antibiotics application should be banned in the feed premixes at on-farm for the purpose of growth promotion or as prophylactic. Wastage of feed will lead to reduce the water quality. Good feeding strategies with check trays would improve the good Feed Conversion Ratio (FCR).

4.4.8.6 Water quality

Optimal water quality conditions are to be maintained in the pond. Application of chemicals that contain traces of banned antibiotics to be stopped. Only approved chemicals with proper labeling to be used in need. The optimal conditions mentioned by the MPEDA to be taken as critical limits for maintaining good water quality standards. As most of the farmers indicated that testing of water parameters was taken place for every 30 days. It is essential to test all the water quality parameters every week or at least once in a fortnight. The deviation in change of any water quality would influence the possibility of application of chemicals or antibiotics.

4.4.8.7 Application of Antibiotics

There is an existing practice of using the antibiotics as prophylactic at regular intervals. Application of any antibiotic either banned or permitted for prophylactic purposes should be completely stopped. As the feed technicians were the major source of technical advises for the farmers, the source of technical advice to be revised. The critical limits to be fixed for the usage of chemicals or antibiotics as a regular management practice. As most of the farmers expressed their ignorance about the MPEDA guidelines and banned antibiotics, it is necessary to educate the farmers about the banned antibiotics.

4.4.8.8 Farm Management Practices

The best aquaculture practices fixed by the MPEDA to be implemented in managing the regular shrimp culture practices. It is essential to maintain good hygienic and sanitary conditions in the pond environment. Regular handling of shrimp to be avoided as it may leads to transfer of human pathogen and would in turn lead to the application of medication. Essential care should be taken to control the contamination of any banned antibiotics in the water column, feed and shrimp. For controlling the viral, bacterial, fungal, protozoan or any other diseases, proper diagnosis to be done before applying the medication. Improper diagnosis would lead to the wastage of drug or

chemical to treat particular pathogen. The medication should only contain permitted antibiotics and should not be applied prohibited antibiotics or drugs

4.4.8.9 Harvesting

During the last days of culture and prior to harvesting, the farmers in India were more prone to use squids, snails and other organisms for developing good average body weight. There is a more possibility of entering the banned antibiotic residues from the supplementary feeding other than commercial feeding. The supplementary feeding like use of live organism should be stopped or to be analyzed for the banned antibiotic residues. At the time of harvesting time if the shrimp contain any antibiotic residues, the culture period to be extended for a period of two weeks depending on the concentration of antibiotic for the withdrawal of the antibiotic. The results of withdrawal time for oxytetracycline showed that 16 days were the elimination of the antibiotic from the body of the shrimps. But it was advised for 21 days of time for withdrawal of any antibiotic under given conditions.

4.4.9 Monitoring procedures

For each critical control points, the periodic monitoring for banned antibiotic residues were established (Table.23). As most of the farmers were testing the water quality parameters for every 30 days or 15 days. None of the farmer is testing the shrimp for antibiotic residues during grow-out operations due to non-availability of antibiotic residue testing facilities and kits. It is essential to test the sample of water, feed, shrimps for antibiotic residues for every 30 days and prior to 15 days of proposed harvesting.

4.4.10 Corrective Actions

If any critical control point deviates from the prescribed critical limits, it is essential to correct the CCP in order to bring it to the normal. The corrective actions include delaying of harvest and taking the advice of concerned expert for bringing CCP to normal (Table 24).

Table 24. Identification of Critical Control Points (CCP's) in Shrimp Farming

Sl. No	Major Steps in Shrimp Farming	Is there BAR occur at this step?	Whether preventive measures occurs for BAR?	Is the step designed to eliminate or reduce the BAR?	CCP identified
1	Site selection	Yes	Yes	Yes	Yes
2	Pond preparation	Yes	Yes	Yes	Yes
3	Water filling	Yes	Yes	Yes	Yes
4	Shrimp seed	Yes	Yes	Yes	Yes
5	Feed	Yes	Yes	Yes	Yes
6	Water Quality	Yes	Yes	Yes	Yes
7	Application of antibiotics	Yes	Yes	Yes	Yes
8	Farm Management practices	Yes	Yes	Yes	Yes
9	Harvesting	Yes	Yes	Yes	Yes

BAR: Banned Antibiotic Residues

Table 25. Control Measures and Monitoring of Identified Critical Control Point (CCP's) in Shrimp Farming

S.No	Critical Control Point (CCP)	Control measures	Monitoring			
			What	How	Who	Frequency
1	Site Selection	Follow Proper site selection criteria. Soil treatment and hygienic control. proper site selection in order to prevent BAR contamination from other agricultural and fish/shrimp farms	Shrimp pond Soil	Soil testing	Trained Farm manager (or) Farmer	At the time of site selection
2	Pond Preparation	Proper sun drying the pond bottom soil. Use only permitted chemicals with label.	BAR and Chemicals	Observation for labeling	Trained Farm manager (or) Farmer	Once at the time of pond preparation during each crop
3	Water Source	Water quality treatment prior to seed stocking	Water quality, pathogens, and BAR	Water quality testing	Trained Farm manager (or) Farmer	Weekly
4	Shrimp Seed	Reliable PCR tested seed accompanied with certificate	Post larvae	PCR testing	Qualified laboratory Technician	Once at the time of purchase of seed
5	Feed	Use good quality feed containing good texture	Feed contamination for pathogens,	Feed quality and BAR testing	Qualified laboratory Technician	For each batch of Feed

		and quality	BAR			
6	Water	Water quality treatment and farm hygiene control	Water quality	Testing water for physico-chemical parameters	Trained Farm manager (or) Farmer	Every week
7	Application of Antibiotics	Prohibit using Antibiotics	BAR	Frequent Testing of shrimp, feed, and water for BAR	Trained Farm manager (or) Farmer	Every fortnight and two weeks before harvesting
8	Farm Management practices	Best management practices (or) Standard Operating Procedures, controlling bird menace	Sanitation and hygiene	Testing feed, water samples for pathogens	Farm manager	Every month
9	Harvesting	Apply appropriate withdrawal period(if necessary)	BAR	Testing shrimp	Farm manager	Every month and 14 days prior to harvesting

* BAR – Banned Antibiotic Residues

Many problems can be avoided by taking corrective action in advance such as water exchange and feed adjustment as soon as the first unusual observations are recorded

The survey results showed that most of the farmers were exchanging the water for every month at about 10-30% of total water.. It is essential not to exchange the water unless situation compels and without the advise of expert (HACCP team). If corrective actions suggest the water exchange, it is essential to inform the neighboring farmer about the discharge. Water to be exchanged to reduce phytoplankton, organic matter, toxic ammonia loads and pH in the pond. The feeding strategies to be revised and feed to be if necessary. Avoid completely the use of antibiotics.

If site selection process indicates the contamination of soil with traces of antibiotic residues, it is necessary to make appropriate alternatives or solutions as suggested by MPEDA. The corrective actions include delaying of stocking the post-larvae.

If the PCR tests of post-larvae reveals positive, discard the particular batch of post-larvae. It is appropriate to stock only Specific pathogen free (SPF) post-larvae (if available) in the shrimp ponds for better results. It is advisable to stock the optional number of post-larvae per square meter as per the specifications given by the Government organizations.

As majority of the farmers were using commercial shrimp feeds, it is essential use the feed having good water stability combining with good pellet/crumble texture. Discard the feed, which reported contamination of antibiotics, mycotoxins and human pathogens. Feeding strategies to be reviewed in order to reduce the wastage of feed. It will enable the increasing of good water quality and bottom soil. Farmers should also observe feed waste at the feeding areas and pond bottom under feed sampling trays. In general, shrimp prefer not to feed on polluted areas and thus move to feed in sampling trays instead. This causes the over-estimation of shrimp biomass in pond and subsequently miscalculation of feed requirements.

All the water quality parameters to be maintained at optimal levels in line with the standards of the MPEDA. Good water quality enables good health for the shrimps. If any deviation occurs in maintaining the optimal conditions, necessary control measures suggested by MPEDA to be taken into consideration.

Farm management practices consists of health management practices that includes separation of infected animals and medications as per the instructions specified on the label and the guidelines of MPEDA and USFDA to be taken into consideration for preventing antibiotic residues in the shrimp tissues. Proper withdrawal time to be strictly followed for the complete elimination of the used drug or chemical.

The Farm management practices should reflect the good hygiene and sanitary conditions. It is essential to bring the situation normal if any deviation occurs in maintaining good hygienic and sanitary conditions at farm sites. The pond workers are required to take appropriate precautions while handling the shrimp and feed. The standard operation practices (SOP's) would enable to bring the situation normal in a stipulated time.

If the monitoring reveals the presence of any banned antibiotic residues and antibiotics with MRL's, the harvesting of the shrimp to be delayed by observing proper withdrawal period. The supplement of any feeding other than commercial feed to be stopped. The method of harvesting procedure also to be corrected as per the specification of MPEDA.

4.4.11 Verification

The verification procedures include verifying the individual critical control point and overall HACCP Plan. Periodic verification helps verify success of implementation and improve the plan by exposing and strengthening weakness in the system and eliminating unnecessary or ineffective control measures.

The verification should be conducted periodically both every 30 days and 15 days prior to harvesting by the HACCP team. The team of HACCP can

conduct the verification process. The help of government organizations could be taken for the verification of success of HACCP plan. Prior to 15 days of harvesting, weekly verification would be more effective for the effective prevention of antibiotic residues.

The verification activities include :

- (a) Verification of effectiveness of preventive control at each CCP.
- (b) Audit implementation (verify effectiveness of the implementation)
- (d) Targeted sample collection and testing

4.4.11.1 Verification of effectiveness of preventive control at each CCP

- (a) Take samples of water, feed, soil necessary and shrimps for banned antibiotic residues testing.
- (b) Take water and feed samples for quality analysis by laboratory
- (c) Take feed samples to check pathogenic bacteria.
- (d) Review extended period of record.

4.4.11.2 Auditing the implementation (verify the effectiveness of implementation)

- (a) Onsite inspection of HACCP related activities.
- (b) Activities conducted as planned.
- (c) HACCP activities that are followed at farm are effective and appropriate.
- (d) Affected products are controlled corrective actions are taken when there were deviations.

There are many activities to be done in order to verify the success of HACCP; therefore, verification plan is needed. As most of Indian shrimp farms are small scale and marginal farms, it would be difficult to operate the verification process on their own. The education background would also play major role as most of the shrimp farmers are having the qualification of

matriculation (SSC) or Higher Secondary (HSC). Very few farmers are professionally qualified. In operations where capacity and resources are limited, a well-planned verification program is essential.

4.4.12 Documentation

In order to implement HACCP at shrimp farms, the farmers must maintain records of all CCP monitoring activities, including records of feed, seed, water sources, chemicals, antibiotics, drugs, production data and monitoring activities.

4.4.12.1 General Pond Data

It includes pond size, quantity and date of seed stocking should be recorded. The number of seed should be estimated after arrival at the farm otherwise mortality during transportation may not be accounted for, leading to over estimate of feeding requirement and shrimp survival

4.4.12.2 Water quality

The important parameters of water quality, which should be recorded in the morning and afternoon each day, include pH, dissolved oxygen and turbidity. Alkalinity, ammonia, nitrite and temperature should be recorded when there is weather change (i.e. heavy rain, sudden warm or cold spell) or at least once a week. Regular monitoring of dissolved oxygen can help optimize use of aerators by determining the period when they are actually required, thus reducing power costs. Continuous monitoring of water quality parameters and study of their correlations will help the farmers to understand their pond ecosystem, and to subsequently forecast environmental changes and take precautionary measures to prevent problems.

4.4.12.3 Feed

Feed weight per meal and per day as well as accumulated feed should be recorded daily and compiled after every sampling of body weight in order to check feeding efficiency. Calculation of FCR from these records is also used for monitoring feed management. Monitoring of feed consumption by

feed sampling trays should be regularly practiced and clearly recorded so that serious problems be identified immediately. If feed consumption is decreasing in the afternoon, farmer will be alerted and may be able to identify its major cause by examining recent records of water quality. A decrease in shrimp feeding may be caused by extremely high pH and warm water, which accelerates the toxicity of unionized ammonia.

4.4.12.4 Shrimp Growth

Regular sampling of shrimp body weight should be carried out every 7-10 days in order to estimate shrimp biomass in the pond. This shrimp growth monitoring is also useful for comparison with standard growth; for adjustment of feed quantities; and for making decisions on harvesting.

4.4.12.5 Production Cost

If production cost has been regularly recorded, a farmer will be able to update his benefit / loss, which is used for decision making on harvesting or extending culture period to receive optimal benefit.

The collection and monitoring of this data assists in carrying out verification activities. This intern will help in reducing the complicit antibiotic residues from shrimp farms. During the survey it was observed that majority (90%) of the farmers were not maintaining any records about the details mentioned above. Only few farmers who possess education of graduation level were have information in papers but lacks proper data and other details required for HACCP documentation procedures.

Table 26. HACCP Plan for Indian Shrimp Farming to Prevent Antibiotic Residues

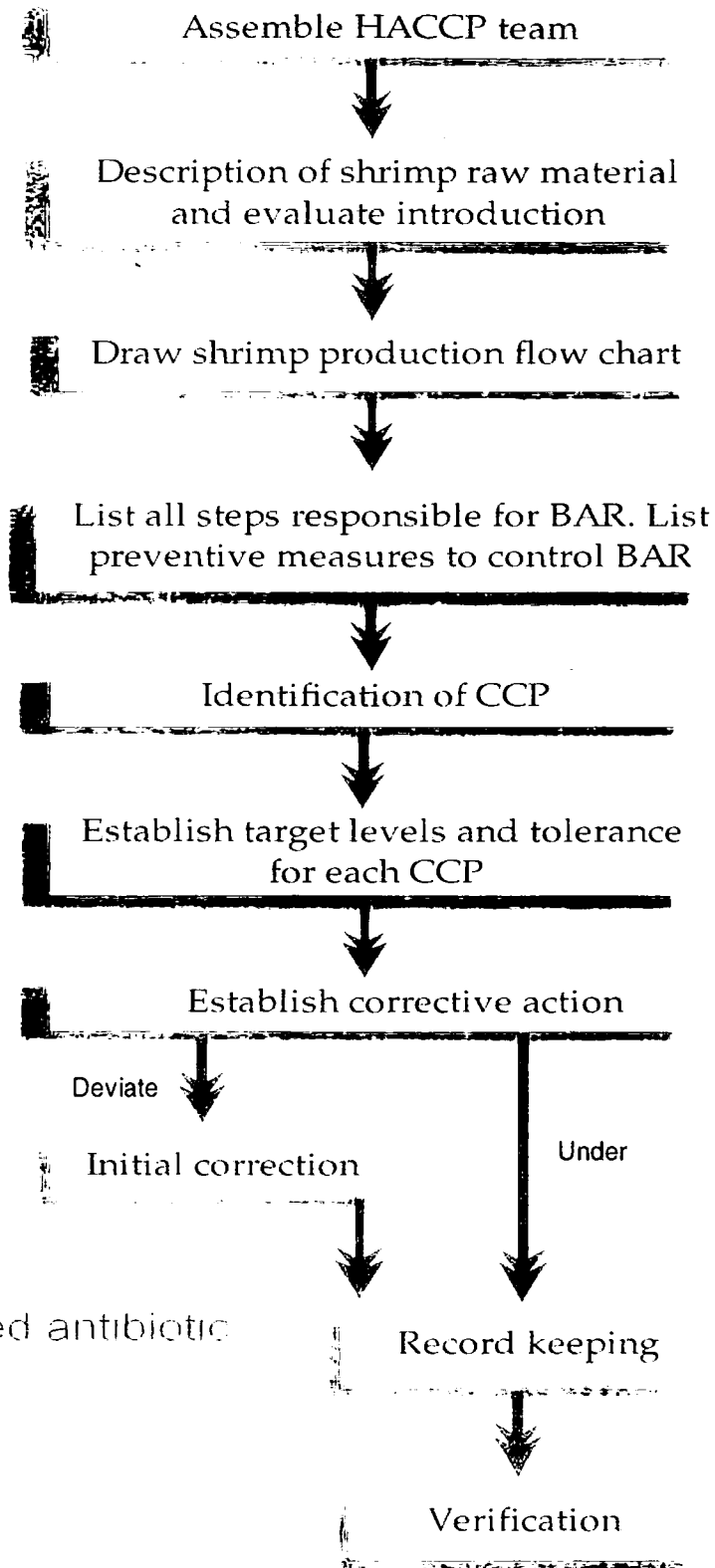
Shrimp Farm _____ Date _____ Page _____

S. No	Critical Control Point	Hazard	Control Measures	Monitoring Procedures	Monitoring Frequency	Specifications	Corrective Actions	Person Responsible	Record Keeping
1	Site selection	BAR	Proper site selection criteria. Soil treatment and hygiene control.	Soil testing	Once at the time of site selection	Acceptable according to specifications of MPEDA	Soil treatment	Farm Manager (or) Farmer	Farm log book, farm hygiene record, site selection record
2	Pond preparation	Contamination of BAR	Use only labeled chemicals for pond treatment, sun dry the pond	Application of chemicals and other pond preparation activities	Once at the time of pond preparation for each crop	Labeled chemicals and fertilizers not contaminated with BAR	Discard the unlabeled products. Sun dry the pond bottom and give sufficient time before stocking water	Farm Manager (or) Farmer	Farm log book - Chemicals and fertilizer records - pond preparation record

BAR – Banned Antibiotic Residues

S. No	Critical Control Point	Hazard	Control Measures	Monitoring Procedures	Monitoring Frequency	Specifications	Corrective Actions	Person Responsible	Record Keeping
3	Water source	Contamination of pathogen -s and BAR	Source of water quality and water treatment	Water quality and contamination of BAR	Each time during water exchange	Free from disease pathogens and BAR	Re evaluate the source of water, retain water at longer period in treatment pond before use	Farm Manager (or) Farmer	Farm log book -Farm hygiene -Treatment of water
4	Shrimp seed	BAR, and Viral pathogens,	Reliable PCR testing	Monitoring of seed survivability	Once prior to seed stocking	Test results should be negative	Discard the complete batch	Farm Manager (or) Farmer	Farm log book - seed details - PCR results - other tests
5	Feed	Contamination of BAR and pathogenic bacteria	Farm hygiene control, Feed storage, prohibit mixing of Antibiotics	Purchasing of feed and on farm	Once in a month and 2 times prior to harvesting	Feed contamination pathogenic bacteria and BAR not reported	Discard the contaminated feed	Farm Manager (or) Farmer	Farm log book - Feed details - Test results

S. No	Critical Control Point	Hazard	Control Measures	Monitoring Procedures	Monitoring Frequency	Specifications	Corrective Actions	Person Responsible	Record Keeping
5	Water	Contamination of BAR	Farm hygiene control, water treatment	Monitor water quality	Every week	Acceptable according to MPEDA specification	Water treatment	Farm Manager (or) Farmer	Farm record -Water quality
7	Application of Antibiotics	BAR	Prohibit usage of antibiotics	Monitor the labeling and banned antibiotics	Every week	Acceptable according to MPEDA and GOI specifications	Delay the harvest, discard the contaminated product	Farm Manager (or) Farmer	Farm record -Antibiotics, drugs, chemicals, etc.
8	Farm management practices	Antibiotics	Best Farm management practices (or) standard operating procedures	Monitor water, feed and shrimp quality	Every fortnight and weekly prior to harvesting	—	Treatment of diseases and improvement of sanitation at farm locations	Farm Manager (or) Farmer	Farm record: -farming practices
9	Harvesting	BAR and residue exceeding acceptable level	Apply proper withdrawal period	Review records of antibiotics, drugs or chemicals application	Every week throughout culture period	—	Appropriate withdrawal period, send raw material for BAR check up 14 days prior to harvesting	Farm Manager (or) Farmer	Farm record book. -Record details of application Antibiotics, drugs and chemicals



*BAR-banned antibiotic residues

Fig. 9 HACCP framework applicable at shrimp farms in India to prevent antibiotic residues



Discussion

5. DISCUSSION

5.1 FIELD SURVEY AT COASTAL SHRIMP FARMS OF INDIA

5.1.1 Profile of Characteristics of Shrimp Farmers and Farms

The results pertaining to the profile of the shrimp farmers and farms showed an over all similar characteristics with respect to the east coast and west coast regions. Majority of the shrimp farmers possess etc. above the primary education level and performing only aquaculture activities for their livelihood. All the shrimp farmers were having minimum three years of experience in shrimp farming. The results pertaining to farm type showed varied results for east coast and West coast regions, as majority of the east coast farmers performing shrimp farming activities in their own farms and the West coast farmers were carrying farming activities in leased farms. The variation of the results with respect to farms type might be due to the sample size and location. The results pertaining to soil texture showed that the majority of the shrimp farmers were culturing in sandy loamy type soils. Majority of the growers possess 0-2 ha q farm area.

According to MPEDA (1996), Upadhyay (1994) and Boyd (1998), an ideal soil texture for construction water retention are clay loam or loam. Many coastal soils are usually high in sand content and will not hold water. The potential soils must have reasonable clay content (particle size of less than 0.002 mm) so that the pond can hold water. Sandy clay or Sandy loam types of soils are best for dike construction, as these are hard and do not crack on drying. Most of the shrimp farms in Andhra Pradesh especially the farmers in Nellore district were having the seepage problem due to poor soil texture. It is essential to follow the proper site selection criteria for construction of new farms and the existing farms should make up the farms by keeping reasonable clay content as specified above. The seepage problem due to poor soil would lead to more utilization of electricity consumption and thereby increasing cost of production for each crop. According to Anon (2005b),

shrimp production in Andhra Pradesh costs around Rs.120/- per kg for 40 count tiger shrimp and Rs.160 per Kg for 30 count. The survey results are agreeing that the more than 90% of shrimp farmers in the county belong to small-scale farmers (0-2 ha). In Andhra Pradesh, farmers owing less than 2 ha dominant (96.18%) while in Tamil Nadu 16% of the farmers are less than 2 ha (AAI, 2001; Vasudevappa and Seenappa, 2002).

5.1.2 Characteristics of Intensity of Shrimp Farming

Majority of the shrimp farmers were taking 2 crops per year. The stocking was generally carried out during the months of Jan/Feb for first crop (Kharif season) and June/July for the second crop (Rabi season). Most of the farmers were harvesting after 120 days culture period. The farmers who were taking more than 2 crops might be due to condensation of harvesting period i.e. less than 120 days or due to attaining the marketable size of shrimps before that period. It can also be inferred that the farmers who were taking 2 crops per year might be not following crop holiday between the crops. In general, October to December period would be treated as crop holidays. The usage of aerators would enhance the problem of Dissolved oxygen (DO) in the pond. The results showed that few of the West coast farmers were not using aerators. It might be due to the availability of optimal DO conditions in that pond or the farmers might be unable to bear the expenses of aerators. The results indicated that majority of the Indian shrimp farmers were performing water exchange either for every month or fortnight. It reflects the possibility of occurrence of hazardous substances that pose the problem of diseases or mortality. Some farmers in west coast region of India were not performing water exchange throughout the culture period. It might be due to the maintenance of optimal conditions in the pond water or might be due to the water scarcity. The results also indicated that most of the farmers growing shrimps in their own ponds throughout the year. It could be due to either difficulty in maintaining the crop rotation or due to fear of problems associated with the crop rotation practices. Most of the farmers in Kerala were performing crop rotation. It was due to traditional shrimp farming practices where crop rotation would be carried either with paddy or shrimp at regular intervals.

According to MPEDA (1996), and Boyd (1998), oxygen is considered as the prime limiting factor in the aquatic environment while 21% is the oxygen content in atmosphere, depending on salinity and temperature. Water normally contains 4-14 mg/lit of dissolved oxygen. Dissolved oxygen concentration is one of the most critical parameter affecting growth in shrimp farms. High dissolved oxygen levels in aquaculture system are to be maintained for increasing production per unit area. Aerators are the energy inputs to improve oxygen transfer in the water surface. They mix the water, so that areas of low oxygen are brought in contact with atmosphere. It is essential to reduce the water exchange as far as possible by keeping good culture conditions.

The results pertaining to stocking density of shrimp farms showed that most of the farmers stocking 5-12 PL's per square meter. The source of water used for the shrimp culture was creek water and bore well. Majority of the west coast farmers were using the creek water and the east coast farmers were using either bore well or creek. According to AAI (2001), Shrimp farming in most of the states are creek based. Like other water sources, surface waters, brackish and seawater sources are subject to contamination usually increasing in severity the closer one is to the shoreline. Coastal pollution in an even worse without strict governmental regulation. Water intakes should be located as far offshore as economically practical and should not be located near industrial or municipal discharges or near area subject to agricultural runoff. Salt-water intrusion in some coastal regions makes it possible to extract brackish water from the ground. The quality of such well depends on the site conditions and geology of the water bearing strata. Typically, the deeper the well, the more expensive the construction and pumping costs. A disadvantage of salt wells is that site conditions and geology often produce unfavorable ground water quality (Chandrakant, 2004; Upadhyaya, 1994 and MPEDA, 1996).

5.1.3 Shrimp farm management practices

Most of the farmers were considering the importance of water quality in their farm management practices. The water quality parameter includes pH,

salinity, transparency, dissolved oxygen water colour etc. The maintenance of plankton in the pond water was also treated as one of the important aspect to be considered for good water colour. The results showed that farmers from all over India were maintaining optimal pH in shrimp ponds. The results pertaining to the salinity indicated the presence of low saline conditions in the water used for shrimp farming. Majority of farmers from Andhra Pradesh especially Nellore district were growing the shrimps at zero saline waters. It might be due to the fact that virulence of white spot syndrome virus was less compared to high saline waters (Anon, 2004; Vasudevappa and Senappa 2002).

Most of the shrimp farmers in India were keeping optimal water transparency in their shrimp ponds. The results indicated that majority of shrimp farmers experiencing the problem of poor plankton growth in their ponds. Farmers from Gujarat state reported optimal plankton growth and this was attributed to the availability of good soil and adopting good farm management practices. According to Upadhyay (1994) and Boyd (1998), turbidity in pond water may be that resulting from phytoplankton blooms and that caused by soil particles. The more turbid the water the less the light penetration into the pond water. Less light at pond bottom discourages the growth of filamentous algae grow on the bottom (benthic algae) and provide natural food for fish. However, in many cases, phytoplankton bloom is preferred than growth of algae at the pond bottom.

The survey findings showed that a few farmers were not testing any of the water quality parameters. This could be due to non-availability of water testing kits and lack of sufficient knowledge on water quality management. The results indicated that most of the shrimp growers were more than 1m water depth. Optimal water depth would enhance optimal growth of the shrimp.

5.1.4 Sources of Seed and Feed

All the shrimp farmers in the surveyed states were stocking hatchery seed in the grow-out ponds. Even though the traditional farming practices

were carried out in Kerala and Karnataka (Vasudevappa and Seenappa, 2002), the present survey indicated that the farmers were stocking the hatchery produced seed. This could be due to adaptation of scientific farming for better crop yields. The results indicated that some farmers (11.7%) were using PL attained the age of more than 20 days. Keeping in view of the survivability PL, it would be essential to stock the PL of 15-20 days.

The findings pertaining to the feed usage revealed that a few number of farmers were using farm made feeds and majority of them were using commercial shrimp feeds. The survey findings also revealed that about half of the Indian shrimp farmers were not obtaining good feed conversion ratio. The results are agreeing with that of Anon, (2004). In order to bring down the cost of production for the shrimps under the prevailing conditions, it is necessary to the farmers to give attention towards FCR. The present FCR of 1.8 should be brought down to 1.5. As shrimp prices were good till recently, farmers did not bother about the feed consumption and its cost. Since prices for the end product (shrimp) have come down drastically farmers should focus their attention on FCR and work towards, enhancing their efficiency. Every point of reduction in FCR will save Rs.5000 per ton of shrimp to the farmer. FCR of 1.5 is acceptable in shrimp culture. For one lakh pieces of shrimp, the farmer can save Rs 50,000 with 1.5 FCR. In kilogram terms, the saving would be Rs.15, a Kilo and Rs.15,000 per ton, on feed alone.

Few farmers in west-cost region were using both commercial and local or farm-made feeds. It is known that aquaculture feeds that contain higher levels of moisture are prone to faster microbial spoilage due to the action of various bacterial enzymes (Jones, 1987) Raghavan, (2003), reported that the farm-made feeds analyzed showed a higher bacterial load of 10^6 to 10^7 cfu g^{-1} indicating the advent of bacterial contamination. Most of the farms made feeds analyzed were wet feeds prepared from animal-based raw materials such as prawn meal, shrimp-head meal, fish meal and clean meat which usually contain very high levels of moisture. The farm-made feeds prepared from marine animal-based raw materials, especially those which are

commonly used in traditional shrimp farms, are in most cases contaminated with various species of human pathogenic bacteria.

The survey finding showed that half of the growers were performing PCR tests prior to stocking and few farmers were not performing any tests on shrimp seed. This is leading to the problem of white spot syndrome virus in the grow out ponds. According SIFT (2004), 310 samples yielded positive results for white spot syndrome virus of total 1232 samples collected from shrimp ponds in East and West Godavari districts of Andhra Pradesh. The report also indicated the increase of positive samples of WSSV from 2001 onwards. It can be infer that WSSV disease is prevailing in most of the shrimp ponds and might be entering into the grow out ponds through the post larvae. Establishment of commercial hatcheries availing overseas technology paved the way for rapid progress of shrimp farming in the country (Vasudevappa and Seenappa, 2002).

5.1.5 Antibiotics and Chemicals used in Shrimp Farming and their Relation with Other Variables

The results pertaining to the usage of antibiotics in the shrimp farming indicated that most of the farmers were not responded and did not reveal any information. The correlation coefficient results indicated that the possibility of using antibiotics by the non responded farmers. A few farmers only revealed the usage of antibiotics. The correlation coefficient results indicated the positive and significant relationship between the usage of probiotics and education stocking density, culture conditions and water source. Farmers who were having higher education back ground and having more stocking density had been using probiotics in order to maintain good pond environment and the improve the health condition of the shrimps.

The results pertaining to the usage of molt inducers, sanitizers probiotics profile of characteristics of shrimp farmer and farms, stocking density showed that majority of the farmers were using probiotics in the shrimp farming operations. As majority of the farmers were not responded and

did not reveal the use of antibiotics in the shrimp farms. Anon, (2005a) reported the presence of banned antibiotic residues in the harvested raw material of Indian consignments and the Japanese authorities found traces of banned antibiotics in four containers of shrimps. The European Union countries had stopped import of Indian shrimps complaining of traces of banned antibiotics. Most of the chemicals such as sanitizer molt inducers etc were not property labeled and not sure about the contamination of the banned antibiotics. The indiscriminate use of antibiotics, drugs, and chemicals could be largely due to strong marketing strategy adopted by the drugs and chemicals manufacturing companies. These products, mainly available for human or veterinary uses, are sold to gnillable farmers to enhance the productivity of the water bodies, increase resistance in shrimps, promote growth and also protect against diseases such as white spot virus etc .The national survey conducted under the ADB/NACA sponsored regional study on aquaculture sustainability and environment included 966 shrimp farmers spread over four states in India, indicated the usage of several drugs antibiotics. and chemicals to treat the disease out breaks. The causes of disease were unknown to farmers, and no reports successful control measures. The great diversity of chemicals used by farmers and the limited success achieved reflects a need for proper extension service to educate the farmers regarding disease identification, prophylaxis and control (Pathak and Palaniswamy, 1995; Pathak et al., 1996). There were no control exists on the use of feed additives used in shrimp farming. In the present study, findings revealed that more number of farmers were using feed and water probiotics of different brands. The more use of probiotics will incur an additional expenditure of cost of production. Saju *et al.*, (1995) reported that shrimp feed constituted the largest part of variable cost (53.6%) followed by cost of seed, pumping expense, pond preparation, harvesting expenses and sampling expenses. As at present, the cost of chemicals is occupying the next place of cost of seed. It is necessary to check the bacterial count mentioned on the label in order to avoid indiscriminate use of probiotics having lesser the number of bacterial strains. There measures in turn would help the farmer to reduce the cost of production.

5.1.6 Characteristics of Marketing

There were some Indian farmers especially farmers from Andhra Pradesh were selling harvested raw material through the agents or middlemen. It is essential to sell the material directly to the company people or repetitive of processing plants or members of Seafood Export Association of India (SEAI). Appropriate precautions are to be taken to avoid contamination with antibiotics. Prior to harvesting, samples from the shrimp farms to be tested for the antibiotic residues. As microbial hazards were reported (Anon, 2003) due to ice used for chilling the harvested shrimp, precautions are to be taken to avoid any contamination before reaching the processing plant or market. Marketing shrimp raw material as 'head-on' type is always feasible to avoid any pathogenic microbial growth.

5.1.7 Technical Awareness on Shrimp Farming

Prior to starting their own farming, majority of the farmers from the states of Gujarat and Maharashtra had undergone training on shrimp farming. These can be attributed to the successful shrimp farming compared to other east coast states where the problem of antibiotic residues was more predominant. Keeping in view of the successful farming in the states of Gujarat and Maharashtra, it can be inferred that training would help to minimize the problems in shrimp farming. The results pertaining to the awareness of banned antibiotics majority of the farmers indicated their knowledge about the banned antibiotics and a few farmers only indicated their ignorance. Despite the awareness on banned antibiotics, shrimp farmer use antibiotics in their anxiety to save their aquatic crop from attacks of bacteria (Anon, 2005a). Until the time that all farmers are adopting Good Aquaculture Practices (GAP's), there is urgent need for intensifying the campaign against the abuse of antibiotics and to appraise its consequences to the trade and possible impact on future existence of aqua farms.

Most of the shrimp farmers had expressed their unawareness of MPEDA guidelines with respect to sustainable shrimp farming indicates the lesser impact of the MPEDA campaign about sustainable shrimp farming

practices without using antibiotics. The awareness programs to be intensified further to reach the information to all the shrimp growers. The extension programs in this aspect to be evaluated further and modified according to the farmer's need so as to reflect prevailing situation at the national and international markets.

5.1.8 Problems in Shrimp Farming, Reasons for Crop Loss and Measures Suggested by the Farmers for Better Results

Most of the farmers reported the severity of the viral and bacterial disease. The white spot disease was the main reason for several crop losses. Most of the farmers indicated the problem pertaining to the low market price, loose shell, seed quality, water quality, etc. Majority of the farmers suggested the best aquaculture/farm management practices would yield better results in the shrimp farming. Farmers were in need of subsidies and insurance for their farming activities in order to sustain the shrimp farming activities. These measures in turn might encourage farmers to adopt HACCP standards at farm level, which will solve most of the food safety and quality issues.

The National consultation on shrimp farming, a one-day national meet held at Chennai on May 25th, 2005 organized by National Commission on farmers, Aquaculture Authority and the Government of Tamil Nadu, had also revealed the above mentioned problems (Anon, 2005b). Quality seed stock for shrimp farming, attracting bank support and insurance, recognizing shrimp farming in land use classification and uniform lease policies were some of the issues discussed in the meet. In spite of the significant developments made in disease diagnostics, the white spot virus continues to affect the shrimp farms. This had led to successive crop failures and economic hardships. The lack of alternative forms of aquaculture to utilize the shrimp ponds in the coastal areas has further aggregated the problem. Of all the issues impacting sustainable shrimp farming in the country, disease and health management issues are the most pertinent. In the absence of an assured source of brood stock, the availability of quality seed is also likely to be major problem in the coming years. As most of the farmers indicated the electricity problems especially in states like Andhra Pradesh, the priority should be given for

electric connection for pumping water in aqua farms as admissible in Agriculture sector. The small and marginal farmers to be provided with subsidy for purchase of shrimp seed and feed. Most of the farmers were having crop damages due to white spot virus, bacterial disease and floods etc. to be suitably compensated through insurance or Government subsidy. Most of the issues relating to the shrimp farming would be addressed once the 'coastal aquaculture authority' bill, which was passed in the parliament on May 5th, 2005, becomes an act. The act would provide for under taking Coastal Regulation Zone (CRZ) and brighter prospects of security loans and obtaining insurance cover of their activity, once the act is on the India statues book (Anon, 2005a).

The other problem, which is causing severe concern in shrimp farming in recent years, is Loose Shell Syndrome (LSS). While the incidence of the problem seemed to be on the increase, no clear understanding of the problem was apparent. A workshop conducted in Chennai on April 28th, 2004 had emphasized the causative factors and discussed in detail. The workshop found that the loose shell problem was estimated to affect between 40 and 50% of farms in 2003. The problem occurs more in summer months (April to June) when the animals are in 60-90 days of culture. The problem typically occurs when the pond water quality is poor, especially when there is a bloom of blue-green algae and Dinoflagellates and high bacterial load. The problem of loose shell can be attributed to inadequate or nutritionally imbalanced feed, infections agents, poor water quality, poor quality seed and pond bottom. In the present study, majority of the farmers suggested the importance of Good Management Practices (GMP) and showed interest in adoption of new technology.

To avoid indiscriminate use of antibiotics in shrimp farms, problems of diseases to be addressed as most of the farmers might be using the antibiotics and other chemicals for preventing the above mentioned problems in their farming practices. It is essential to implement the HACCP framework coupled with Good Management Practices at shrimp farm sites in order to address all the problems in shrimp farming.

5.2. Estimation of Oxytetracycline Withdrawal Period in *Penaeus monodon* Reared in FRP Tanks

The results clearly proved that antibiotic residues in the tissues and a withdrawal period of 15 days. Thus the residue of OTC was totally abolished by the 16th day. Consumption of shrimps within this period would pose human health hazards and hence the need for the setting up of a critical limit. Based on the present results as also earlier studies (MPEDA, 1996), and taking into consideration the fact that many other antibiotics may also be used, a period of 21 days post medication should be enforced as a corrective/control measure. However, it is imperative that the ban on antibiotics is strictly enforced.

5.3 Hazard Analysis of Feed Samples

The microbiological hazard analysis of different commercial feed samples showed that most of the feed samples contain bacterial loads that were within the permissible limits. Even though the microbial contamination was normal, the variations of bacterial load in feed samples indicated the unhygienic handling of the feed by the farm workers and the feed storage conditions at different shrimp farm sites. If any deviation of the existing hygienic and sanitation conditions at shrimp farms could pose the possibility of contamination of human pathogenic bacteria. Raghavan (2003) and Anon (2003), reported similar type of results in the commercial feed samples as total bacterial count ranged between 10^3 - 10^5 cfu/g. Commercial feeds are considered unsatisfactory if they contain large bacterial population even if the bacteria are non pathogenic in nature and have not caused any changes to the pellet (Thatcher and Clerke, 1968).

Modern and hygienic methods of feed processing and improved feed management practices at the farm sites have greatly reduced the incidence of such bacteria in shrimp feeds. In the present study few feed samples showed the contamination with more number of coli forms and *staphylococcus aureus*. It was due to the unhygienic handling practices followed by the workers at the farm sites. Adequate control over the health and hygiene of shrimp feed

handlers and refrigeration (below 5°C) were the suggested measures (Iyer, 2002).

The physical hazards of shrimp feeds showed that most of the feed samples were not contaminated with any physical hazards except the texture of the pellets of feed samples had shown varied results. The feed sample - C contained poor texture and more powder indicating the defects in feed preparation. It was observed that majority of feeds lost the texture as the days of culture extended. It would be feasible to use the good quality feed having pellets/crumbles of uniform size, shape and colour. The presence of powder in the feed shows the poor quality of pelleting (MPEDA, 1996). If shrimps fed with powdered feed, there will be more loss of feed as the dust will be blown up in the wind. Water stability of feed is another important factor to be considered for the good quality feed for grow-out shrimp production. Binders used in the feed provide the desired water stability, to prevent disintegration of feeds and leaching of nutrients into the water (MPEDA, 1997). While some ingredients and additives used in aquaculture feed pose hazards. Mycotoxins are metabolites of fungi of various genera. Such fungi can grow on feed ingredients before and after harvest or during transportation or storage (WHO, 1999).

The quality of the feed depends upon the raw material quality and quantity, processing conditions and shelf life. The efficacy of feed in promoting growth primarily depends on the above aspects. Poor feed quality can be due to (i) poor quality raw material (ii) adulteration with urea (iii) high acid-insoluble ash representing sand and silica (iv) high ash level (v) use of rancid levels (vi) inadequate vitamin levels in premix (vii) low levels of highly un saturated fatty acids (viii) low levels of essential amino acids: imbalance of Calcium Phosphorus ratio (ix) presence of anti nutritional factors (x) old feeds- prolonged storage of feed lowers its nutritional quality (xi) high moisture levels (exceeding 13%) and (xii) high salt content. Storage of feed ingredients and finished feeds (pellets, granules or flakes) is an important step in maintaining the quality of the feeds. Proper storage and handling is necessary to prevent

theft, physical and chemical damage. The accumulated economic loss can be enormous due to the quality changes (MPEDA, 1997 and De Silve, 1998).

The feed sample – C showed the presence of *Escherichia coli* (Plate) is often used as an indicator for faecal contamination. Some strains of *E.coli* are capable of causing food borne disease, ranging from mild enteritis to serious illness and death (WHO 1999). Presence of faecal contaminating bacteria and faecal *streptococcus* and *Staphylococcus* indicates general break down in hygiene and sanitation. Feed contamination may take place through utensil, hands through improper handling. When conditions favorable, the contaminated organisms multiply rapidly and further aggregate the situation. The control measures for faecal contamination include cold storage as 95% reduction of faecal coli form at -4°C (Iyer, 2002).

Commercial feeds are considered unsatisfactory if they contain large bacterial population even if the bacteria are non-pathogenic and have not caused any changes to the pellet. Higher level of moisture is prone to faster microbial spoilage due to the action of various bacterial enzymes (John, 1987).

5.4 Development of HACCP Framework for Indian Shrimp Farming to Prevent Antibiotic Residues

The results indicated that the developed framework had identified nine critical control prints (CCP's) with respect to the Indian conditions. Most of the potential hazards and their causative substances would occur at the identified CCPs. The pre-stocking operations include site selection, pond preparation, and water source. The post stocking operations include shrimp seed, feed, water quality, application of antibiotics, farm management practices and harvesting. The success of application of HACCP principles to control the antibiotic residues depends upon monitoring, corrective actions and record keeping. In the present study feeds were analyzed for microbiological and physical hazards, due to its share in the cost of production. The problem of antibiotic residues is occurring due to on- farm mixing of antibiotics in the feed by most of the growers on a daily basis. In spite of this, there is more possibility of contamination with antibiotics either during manufacture or in the

other stage of distribution. It is essential to perform feed monitoring for antibiotic residues by collecting the feed samples of same batch from the feed plant, feed distributor and farmer.

Garett *et al.*, (1997) had identified pond site selection, water supply quality and pond management techniques as the critical control points for shrimp aquaculture to control the spread of exotic animal viruses. In one study in Vietnam, water supply, fish fry, fish feed and pond conditions were identified to control food borne trematodes in fish ponds. Reilly and Kaferstein (1997), had identified site selection, water supply and feed supply for the aquaculture production. Gunderson and Kinnunen (2001), had developed the HACCP framework by identifying the above critical control points to prevent the spread of aquatic nuisance species in aquaculture and bait fish operations. Jahncke *et al.*, (2001 and 2002) had identified out source-shrimp, water and fertilizers as critical control points to prevent exotic viruses at shrimp production and processing facilities.

The success of the developed framework depends on the successful control of the antibiotic residues at the identified critical control points. The HACCP approach emphasizes process control. It concentrates on the points in the process that are critical to the safety of the product. It requires regulator, hatchery manager, shrimp farm manager, processing plant manager and representatives of Seafood Export Association of India (SEAI) to communicate and work with one another. Processing plant manager have to verify the significant food safety hazards have been properly identified in general and antibiotic residues in particular.

The developed HACCP framework will provide shrimp farmers with a recognized risk management approach to address the issue of antibiotic residues and control the problems of diseases and environment pollution. It will be a model for use in aquaculture industry in India and world where similar problems and geographical condition exists. It helps to reduce the frequency of use and quantity of chemo therapeutic and other chemicals that are used in shrimp culture practices. Apart from that It also helps to protect animal, environmental and public health. As the processors are considering to

conduct on-site audits of the animal drug controls used by the shrimp growers, this HACCP framework would help in reducing the risk reduction and there by help the processor in promoting the product at international market.

From the field survey it was found that feed technicians were the main source of technical knowledge and influencing the regular shrimp farming practices. Therefore, it is necessary that only qualified personnel to be made compulsory for the source of technical help in the shrimp farming operations and to control the indiscriminate use of antibiotics. According to FAO (2002), approved antibiotics can be brought and utilized under two conditions: over the counter, or on prescription by a qualified professional. For extra-label use, a qualified professional may write a prescription for the use of an approved antibiotic under conditions that vary from those approved. In aquaculture, antibiotics are generally administered in feeds, having been either added during feed manufacture or surface-coated on to pellets by the manufacturer or the farmer. During out breaks of disease farmers may apply antibiotics by mixing in the feed or other routes. Clear instructions are therefore required for the feed manufactures, antibiotic dealers, veterinary authorities and farmers who are responsible for the use of antibiotics. All the elements for identifying the Critical Control Points (CCP's) and critical limits of regulatory requirements exists for approved antibiotics and veterinary drugs, specific fish or shrimp, diagnosis (purpose of use), dose duration of treatment and withdrawal period. Feeding stage was one of the CCP identified since this is where antibiotics are usually introduced into the production process. The analysis of residues of the antibiotics used, and the checking of compliance with regulations, would form part of the verification procedures. USFDA(2001) has suggested that the monitoring of residues in flesh may be not enough, and the development of resistance in pond micro-organisms should also be monitored, as an additional CCP.

The proper use of approved antibiotics will continue to be necessary in animal production, including aquaculture, and consumers and importing countries should be reassured the use of approved antibiotics, in particular

under label use conditions, does not imply a hazard. However, the regular and indiscriminate use of chemo-therapeutants leads to several compliances. The routine use of these drugs in aquaculture practices often leads to morphological deformities and mortalities in the host, development of resistant strains and public health hazards (Baticados and Paclibore, 1992). Seafood processors have to depend on the declarations from the shrimp farmers or supplier to the effect that no antibiotics have been used in the farm.

Surendran *et al.*, (2003) suggested that no aquaculture drug to be used in the farm apart from controlling measures taken by Government agencies in addressing the antibiotic residue problems in Indian seafood. Abraham (2002), pointed out lack of regulations on the use of antibiotics in Indian shrimp aquaculture could become as much a threat to the aquatic organisms at the pathogen themselves, and the environment and the humans at large. As suggested by Aquaculture Certification Council (ACC, 2004) of USA, banned antibiotics, drugs and other chemical compounds shall not be used in order to address the issue of food safety. Other therapeutic agents shall be used as directed on product labels for the control of diagnosed diseases or required pond management, not prophylactic purpose. Shrimps shall be periodically monitored for residues. The record keeping should be done for 'each pond' and 'each production cycle' and it would include details like antibiotic and drug use, stocking data, harvest data etc. so as to maintain traceability.

The success of implementation of this HACCP framework depends on the monitoring programmes carried at shrimp farms. The monitoring programmes conducted in shrimp farms in Honduras, USA, and yielded positive results. The farms on the Gulf of Fronsés have monitored water quality at 17 estuaries on a weekly basis for eleven years. A review of the water quality data by Auburn University, USA stated that although the shrimp farm area has grown substantially, no increase in eutrophication of estuaries in Southern Honduras has been found. Honduras farms do not produce shrimps that are unhealthy for humans to consume. They maintained monitoring programmes for antibiotic residues in shrimp products in

accordance with European Union requirements, among the strict in the world. In addition, shrimps are routinely tested for residual pesticides, heavy metals and other chemicals.

Although, the present framework was aimed at preventing antibiotic residues, it can also be used to control other food safety hazards such as pesticides, heavy metal etc by making some modifications to the monitoring programmes. The Department of Fisheries (DOF), Thailand has developed and provides a certification system for quality cultured shrimps to address the issue of antibiotic residues in shrimp (Anon, 2004a). The certification system for Quality Cultured Shrimp is comprised of two systems. Certification system for Good Aquaculture Practice (GAP) will be a certification system for hygienic shrimp hatcheries and farms, and no antibiotic residue in raw materials and Code of Conduct (COC) for sustainable shrimp culture will be a second certification system for a whole production line including hatchery, farm, aquaculture business, transportation and processing. The products must be high quality without antibiotic residues and production process shall be environment friendly. The DOF, Thailand had implemented antibiotic residue monitoring project as antibiotic inspection is employed to detect the presence of prohibited antibiotics. Polymerase Chain Reaction (PCR) is used to detect disease in the brood stock used for shrimp fry production. If viruses are found, the brood-stock can not be used for breeding. Similarly, if there are any viruses in shrimp fry, shrimp fry culture must be avoided. At the shrimp farm, the HACCP team must take care of the shrimp health and if any stress or diseases is noted, shrimp must be sampled for examination and diagnosis. Test kits are the preliminary tests used for the detection of antibiotics in animal feeds. Since quality feed is necessary for the intensive marine shrimp culture the DOF of Thailand carries out feed quality inspections for nutrition, raw material quality and antibiotic presence. DOF certifies shrimp feed by registration of aquatic animal feed. Licenses shall be issued for aquatic feed production and import. Random checking at shrimp feed factory is carried out to ensure the feed quality. HACCP certificate will be issued to feed plant if it meets the required standard. The shrimp feed inspection activity includes efficiency promotion, aquatic feed control, and inspections go to control feed

formula and contamination. Aquatic animal feed samples are collected at the factories, shrimp farms and market for analysis.

For the successful implementation of present HACCP framework, depends on the control measures and monitoring programmes implemented by the Government of India. As the problem of antibiotic residues is still haunting the Indian shrimp farming and the export market, it is essential to implement stringent measures as implemented by Government of Thailand. As of now HACCP is not mandatory for shrimp farms, it is essential to encourage the implementing the HACCP framework at the farm level and to be issued certificates for the farmers who adopt HACCP framework to prevent antibiotic residues. Government agencies like MPEDA, EIC, Aquaculture authority and representatives of shrimp farmer's association, SEAI etc. to be worked with another to address the issue of antibiotic menace.

Selvaraj *et al.*, (2002) suggested that proper withdrawal time must be observed to ensure that the edible tissue is safe when it is offered for sale. The control measures for addressing the antibiotic residues menace including on-farm visits to review drug usage before receipt of the product, regular testing for antibiotic residue, receipt of suppliers lot-to-lot certification for proper drug usage. Pathak *et al.*, (2000) suggested the careful use of antibiotics carefully at correct doses, if necessary. Various chemicals and antibiotics used shrimp farming are mostly available locally, and there is no control on their sale and usage.

It is essential to control the prohibited antibiotics and chemicals by the Government. The good management practices includes stocking disease free and high healthy stock, combine continued assessment of water exchange, use of good quality feed without over feeding, and use sedimentation and recycling ponds (MPEDA, 1996). Preventive health programs at aquaculture facilities include the establishment of a population of healthy, disease-free animals. Good monitoring and record-keeping systems to be implemented. Close surveillance for diseases can provide advance warning of potential problems. Programs can then be established to promote animal health and fight diseases (More *et al.*, 2002; FAO, 2001).

Turbid water may be caused by over blooming of phytoplankton or by siltation after heavy rain. However, rain is unusual in April (according to seasonal weather patterns at the farm location) and always causes water pH to fall.

Shrimp Seed of Quality (SSQ) of Bangladesh, a new technology keeps viruses at far distance by using a closed system, with no water exchange. The program has adopted international code of conduct for the shrimp industry and developed an international credible third party certified quality assurance system to ensure the reputation of Bangladesh shrimp products, to enable them to compete in the global market and fetch premium prices (Anon, 2004). The export-import of Thailand is offering insurance for shrimp exporters against losses incurred if shipments to the European Union are confiscated for chemical contamination. The bank would reimburse the exporter for 70% of the value of the destroyed goods. The production costs would be reduced due to non-use of chemicals, adoption of improved GMP's, yielding better growth, survival and better FCR for increased productivity and returns.

International organizations such as FAO, WHO, OIE and a number of national Governments have already raised the issue of irresponsible use of antibiotics in all production sectors, with particular concern for the potential risks to public health. Many Governments around the world have introduced changed or tightened national regulations on the use of antibiotics, in general and within the aquaculture sector (FAO, 2002).

Unity among the farmers, and co-operation among hatchery personnel, farmers, feed and other input suppliers, processors, exporters and concerned Government institutes are the key for the success. Arranging the 'Aquaculture Club' is a promising model for small-scale farms. As the shrimp prices are fluctuating at regular intervals, the quality of the shrimp will fetch more prices in the international market. The project work of MPEDA/NACA facilitated eleven Aqua clubs in 5 states of India (7 Aqua clubs in 6 villages of Andhra Pradesh and 1 Aqua club each in the state of Gujarat, Karnataka, Orissa and Tamil Nadu). Technical assistance was given to 208 farmers of this Aqua clubs to plan the crop and implementation of the BMP's in their farms. Due to

the successful implementation of BMP's the disease prevalence reduced to a significant level (Padiyar, 2005). The organization of farmers in the form of 'Aqua clubs' to be adopted in all shrimp farms in India for the successful implementation of the present developed HACCP framework. An aquaclub is formed by a group of farmers whose farms are closely situated together in a cluster or locality of a village. The club provides a mechanism for farmers to organize themselves in planning and managing their crop activities and to solve their local problems.

Most of the importing countries of seafood are quality conscious. Government can take necessary measures to make mandatory to introduce HACCP from farm level to input agencies, processing unit and so on. Exporters should limit the purchases from licensed farmers having HACCP certificate for their products.

Keeping in view of the international trade perspectives and WTO-SPS agreement, It is essential to adopt the organic farming, as it protects the health of consumers by reducing the overall exposure to toxic chemicals from pesticides that can accumulate in the ground, air, water and food supply. As organic aquaculture does not use toxic and persistent antibiotics and pesticides and also helps to prevent top soil erosion, improves soil fertility, protects ground water and conserves energy (Diwan and Ayyappan, 2004).

Today, both the consumers as well as sellers are very much interested in environmentally sound seafood products and assure that these seafood products cause no impact on the environment. Therefore, the demand for organic products is very much high in the growing markets. As MPEDA has already taken up this new venture to promote organic aquaculture in India, most of the critical control points could be monitored systematically with the organic shrimp farming.



Summary

SUMMARY

Aquaculture has developed to become one of the fastest growing food production systems in the world with a growth rate 9.6% per year in the last decade and the bulk of its output currently being produced within developing countries like India. FAO (2003) estimates a total production of 41.9 million tones from aquaculture sources, which is approximately 32% of the total fish production. The major increase in global aquaculture production in the last decade has been witnessed by China, India and South East Asia using systems that have already proven sustainable. India is the 5th top most cultured shrimp producer and in the fast few decades, it has become a leading player in both the inland and brackishwater production. Farmed shrimp contributes about 60% by volume and 82% by value of India's total shrimp export. It is one of the thrust sectors of exports and 8th largest foreign exchange earner of the country. Seafood export has grown substantially from a mere Rs.2.46 crore in the early fifties to Rs.6092 crores, equivalent to US\$ 1331 million in 2003-04

India has a long coast line of 8,119 km, a continental shelf of 0.5 million square km, an extensive Exclusive Economic Zone (EEZ) of 2.02 million sq. km, 1.24 million ha of brackish water area, 1,91,024 km length of rivers and canals, 3.15 million ha of reservoirs, 2.25 million ha of flood plain wet lands, 0.29 million ha of estuaries, 1.65 million ha of mangroves, swamps, lagoon etc. All these resources offer immense scope and potential for the development of aquaculture activities. The fisheries sector in India contributed around 1.5% to total Gross Domestic Product (GDP) and around 5% to the GDP from the agriculture sector. Around 7 million people of India are engaged in fisheries and ancillary activities.

Shrimp farming in India has become more popular among the growers. There is 1.2 million ha of brackishwater area spread over 10 maritime states/ Union territories and only 0.15 million ha area is under utilization with an average production of 1 lakh tonnes per year and the projected potential production is 0.3

million tonnes. The production levels which have been steadily increasing reached a total of about 115,000 MT from an approximate area of 1,52,000 ha during 2003-04. Andhra Pradesh continues to witness the maximum growth, followed by West Bengal, Kerala and Tamil Nadu. Presently, the technology adopted ranges from traditional to improved traditional within the coastal Regulation Zone (CRZ) and extensive shrimp farming outside the CRZ. About 91% of the shrimp growers have a holding between 0.2 ha, 6% between 2-5 ha and the remaining 3% have an area of 5 ha and above. Shrimp farming provides direct employment to about 0.3 million people and ancillary units provide employment to 0.6 – 0.7 million people.

About 75% of the shrimp farming area is culturing *Penaeus monodon* being the most preferred shrimp species and because of its high growth and tolerance to environmental changes. It was started in the early 1980's and became a significant commercial activity by 1990 due to the adoption of improved scientific farming. Shrimp production increase showed an impressive growth during 1990-1994, after which production declined due to the diseases outbreaks in late 1994. Crop losses were estimated to be on high as Rs.1,203 crores during the period 1994-98. This disease outbreak led the shrimp growers to indiscriminately use several chemicals and antibiotics. Although these measures did not control the disease out break, it posed another problem of residues of antibiotics in the edible tissues.

With the establishment of WTO in January 1995, quality has become prime factor for exporter in accessing the food markets of the developed world with importing countries imposing strict quality standards. The important markets for India are the European Union, Japan and USA. In spite of MPEDA's ban, usage of several antibiotics continues and the importing countries have been rejecting the Indian consignments due to the presence of banned antibiotic residue and bringing in heavy economic loss. Most of the other competing countries like China, Thailand, Vietnam, Indonesia had upgraded their quality

standards as per the international norms. Apart from the traditionally shrimp farming countries, in the last few years, many other countries such as Brazil, Iran and Madagascar have started producing shrimp.

Unlike other countries, India has vast resources potential for the utilization of brackishwater area and can improve the quality by implementing the Hazard Analysis Critical Control Point (HACCP) at shrimp farm level. In the absence of any systematic study in the direction, the present study had been carried out with the following objectives so as to develop a HACCP framework applicable at shrimp farming facilities of Indian coastal aquaculture.

- To analyse the existing shrimp farming practices in the major shrimp farming states
- To measure level of adoption of scientific farming practices.
- To identify the different sources of hazards leading to the presence of antibiotic residues.
- To estimate the hazardous substances entering through the feed.

The present study was conducted during the year 2003-04 at 120 shrimp farms located in the selected shrimp farming coastal districts of Andhra Pradesh, and Tamil Nadu (East-coast), and Gujarat, Maharashtra, Kamataka and Kerala (West coast). Both purposive and random sampling procedures were used to constitute the appropriate sample for the study.

Feed sample of different feed brands used in the shrimp farms of Andhra Pradesh were analyzed for both microbiological and physical hazards. There were 30 shrimp feed samples of 10 different feed brands were collected from the farmers and were analysed for microbiological and physical hazards. The results showed that majority of feeds possess optimal quality. In order to fix the critical limits and to apply control measures, oxytetracycline a broad-spectrum antibiotic

was used to fix the withdrawal time for *Penaeus monodon* over a period of 90 days.

Medicated feed was prepared by mixing with 5 mg of oxytetracycline per kg of feed and supplemented to animals in experimental tanks at the rate of 3% body weight, for a period of 7 days. The remaining 18 days, non-medicated feed was provided. All the samples were analyzed using High-Pressure Liquid Chromatography using UV detector.

The HACCP frame work was prepared by incorporating the field generated information by using the generic HAACP model to prevent antibiotic residues from shrimp farms

The important research findings of the study are summarized below.

1. The field studies at shrimp farms revealed that majority of the farmers were using the probiotics in order to main good culture conditions. Mainly farmers who were well educated and farmers who stocked more number of shrimps were reportedly using the probiotics.
2. Majority of the Indian shrimp growers possess minimum three years of experience in shrimp farming and have the education level of SSC.
3. Majority of the shrimp farmers were depending solely on shrimp farming for their livelihood
4. Majority of shrimp farms on the east coast were owned by the farmers whereas half of the farms in West coast region were on lease by the farmers.
5. Most of the Indian shrimp farms possess the sandy loamy type soils.
6. Majority of the farmers were practicing two crops per year and resorting to water exchange only once or twice in a month.
7. Most of the water quality parameters were at medium levels.

8. Majority of the growers were not testing the water quality parameters regularly and only half of them were testing it on a monthly basis.
9. All the farmers were stocking the hatchery seed of age of PL 15-20.
10. Majority of the farmers were testing the shrimp seed with PCR for white spot virus.
11. Majority of the growers were using the commercial feeds alone as the feed source.
12. Half of the farmers were obtaining FCR values below 1.5 and the remaining were getting FCR values above 1.5, giving much scope for reduction of cost of production by bringing down the FCR values.
13. Majority of the growers were using the creek water and ground water for stocking the shrimp farms.
14. Most of the farmers avoided to reveal information on the usage of antibiotics. But majority of the farmers were using probiotics, molt inducers and sanitizers.
15. Majority of the farmers selling the harvested raw material directly to the companies and about 42% of the shrimp farmers were depending on the agents or middlemen.
16. Most of the farmers were marketing the cultured shrimp as Head-on and the others were marketing as Head-less giving scope for contamination with human pathogenic bacteria.
17. Majority (71.7%) of the shrimp farmers had not undergone any training prior to starting shrimp farming. But the farmers in Gujarat and Maharashtra had undergone training on basic aspects of shrimp farming which helped them to produce quality seed without using antibiotics.
18. Most of the farmers in India had indicated their willingness to undergo training on the new aspects of shrimp farming with respect to quality aspects. But the few farmers in Karnataka and

- Maharashtra indicated their unwillingness, as they felt that they don't have any problems with respect to quality aspects in shrimp farming.
19. Majority of the farmers indicated their awareness about banned antibiotics. This could be the reason for the farmers not responding to the use of antibiotics in the shrimp farming operation fearing that their product would fetch less value
 20. Mixed results were obtained with respect to the awareness of MPEDA guide lines for sustainable shrimp farming. Majority of Kerala and Gujarat farmers were not aware of the guidelines and nearly half (45%) of the few Andhra Pradesh farmers have also revealed their ignorance about the MPEDA guidelines. This could be the reason for the production of poor quality shrimp.
 21. Majority of the shrimp growers were guided by the concerned feed technicians of the feed used by the farmers. The lack of professional expertise and periodic use of antibiotics and unlabelled chemicals as a regular farming practice could be the reason for the antibiotic residue problem.
 22. Majority of the shrimp farms in India were still suffering from the problem of viral diseases especially White Spot Syndrome Virus (WSSV). Even though most of the farmers stocking the PCR tested seed the bacterial, fungal, and protozoan diseases were still existing in different shrimp farms in India. This could be the reason for the usage of antibiotics and several unlabeled chemicals.
 23. Majority of the problems indicated the problem of low market price, electricity, water quality, lack of lab facility, poor seed quality, subsidies, etc.
 24. Loose shell syndrome was another disease problem reported by majority of the Andhra Pradesh and a few farmers from other states.

25. Majority of the farmers had attributed their earlier crop losses to the diseases especially white spot and poor seed quality.
26. All most all the farmers suggested that best farm management practices would yield the positive results.
27. The hazard analysis of shrimp feed sample indicated that all the feed sample except one are within the permissible limit of bacterial contamination.
28. Half of the collected feed samples indicated the poor texture quality.
29. The results indicated the importance of proper handling of shrimp feed and storage conditions.
30. Pre-stocking operations and post stocking operations, were identified CCP's where the antibiotic residue could be controlled.
31. The 9 critical control points identified under the above 2 broad heads are site selection, pond preparation, stocking water shrimp seed, feed, water quality application of antibiotics farm management practices and harvesting.
32. The critical limits for all the identified CCPs were fixed by using the MPEDA and international standards.
33. Control measures and correction active action were listed out in line with the International standards.
34. The procedures for record keeping and maintenance were illustrated using the standard HACCP generic models.

Implication and Recommendations

1. The framework was developed by incorporating the existing shrimp farming conditions. The framework can therefore be useful tool for Aquaculture Authority, MPEDA, Export Inspection Agency and other Government organizations who play a major role in the aquaculture sector.

2. Though the framework was developed exclusively to prevent the antibiotic residues, but it can also be applied to prevent the diseases and other associated problems. The critical control points would vary as per the target of the framework. It is essential to revise them when the targets of the framework are changed.
3. It is possible to change the present framework as per the existing condition of that individual farm and location, if site-or farm specific requirement arise
4. The framework that was developed in the present study to be tested in demonstration farm in order to reveal the success of the feasibility at small and marginal farms.
5. The developed framework was exclusively prepared for shrimp farming, but the framework can be used as model for scampi and other aquaculture species. This can also be implemented successfully in reservoirs and other big water bodies to prevent problems like turbidity, undesirable and nuisance species, etc. The critical control points would vary according to the culture practice, species and the aim of the framework.
6. The field survey at shrimp farms listed the several problem reasons and suggestion of farmers. The Government can therefore take the survey findings into consideration for the future programs policies and decision-making.
7. Majority of the farmers had indicated their ignorance about the sustainable shrimp farming. It is essential to reevaluate and modify the extension programs in order to make the shrimp growers aware of the latest developments in shrimp culture.

8. The experimental trials of the estimation of the oxytetracycline withdrawal period in *Penaeus monodon* was conducted in Mumbai, Maharashtra state. The other antibiotics have more or less similar withdrawal period for *Penaeus monodon*. So for the safety, the optimal withdrawal period to be considered is 21 days for any given climatic conditions of different locations of India.
9. Feed technicians were the main source of technical guidance for several shrimp farmers in India. The Government should consider the extending the technical expertise to the farmers in the form for providing qualified professionals for the technical expertise.
10. Most of the antibiotics and chemicals used in shrimp farming were not having proper labelling and didn't possess any control over unauthorized selling of banned antibiotics and pharmacologically active substance. Government should make possible efforts in order to control then issues in the form of acts.
11. The survey finding indicated that several farmers were in need of subsidies, insurance to the farm and problems of low market prices. The Government policy makers should consider these issues for the prospects of sustainable shrimp farming in India.
12. Appropriate measures should be taken by the Government to control the problems of diseases, seed quality and water quality.
13. The present study has developed the framework applicable at shrimp farms and the application of this framework has to be extended to hatchery as well as feed plants for effective control of the food safety hazards in shrimp farming.

Suggestions for further research

- a. Further investigations may be taken up in all the districts of all the shrimp farming states to bring out an exhaustive and comprehensive framework.
- b. At present, there is no information on the cost and benefits of application of HACCP principles at shrimp farms. Hence further investigations to be carried by applying the presently developed HACCP framework at demonstration farms.
- c. Most of the investigations including the present study were aimed at health safety of international consumers. Hence further investigations are necessary to analyze the different hazards posing severe health concern of Indian consumers especially rural people including children, by conducting HACCP study in the local fish and shrimp markets and appropriate HACCP framework to be developed for controlling the pathogenic microbiological hazards, Mycotoxins heavy metal, and pesticide residues.
- d. At present there are no studies with respect to the quality of the raw material used in the shrimp feed preparation. Further research has to be carried out to assess for food safety hazards present in the raw material collected from local markets and feed plants.
- e. In the present study oxytetracycline, a broad spectrum antibiotic which is frequently used in hatchery as well as grow-out ponds was used to estimate the withdrawal periods. Further research is needed to assess the effect of different kinds of antibiotics on the environment and development of resistance by bacteria.



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* Not seen in original



Appendix

Questionnaire used for data collection on different aspects of shrimp farming (*Penaeus monodon*) practices

Collected by Sumanth Kumar Kunda, PhD scholar, CIFE, Mumbai.

For use in PhD thesis work (2001-04)

1. GENERAL INFORMATION

1. Name and address of the farmer :

2. Educational background : Literate/primary/HSC/Graduate/professional
3. Occupation : Aquaculture only/Aqua+ Agri/ aqua+ Service (G/P)/Aqua business
4. Farm size :
 - 1) Own/leased
 - 2) Farm area in Ha _____
 - 3) Water spread area in Ha _____
 - 4) Soil type/Texture _____
 Water source: Ground water/surface water (Ground water depth= _____ ft)
5. Shrimp farming experience : _____ years
6. Other than Shrimp farming : Fish culture/scampi culture
7. Do you rotate crop with rice/scampi : Yes / No (if yes mention the type)
8. a) No. of crops per year : ----
9. Seed source & age of PL :
10. Probable months of stocking :
11. Stocking density _____ / M²

II. SHRIMP FARMING PRACTICES

12. Optimum culture conditions
- 1) PH
 - 2) Salinity
 - 3) Temp
 - 4) Turbidity
 - 5) DO
 - 6) Plankton: Poor/Med/Rich
 - 7) Water colour
 - 8) Water depth
13. Pond preparation
- (a) Ploughing Method : Tractor /Bullocks
- (b) Lime application : i) Ag lime/quick lime/dolomite
ii) Dosage :
iii) Frequency application
- C) Manuring : N _____ kg/ha
P _____ kg/ha
K _____ kg/ha
Any other _____
(Vermipost/micro power/Biocult)
PCR test /Stress test /Any other
_____ Days
14. Seed Testing
15. Culture period
16. Aerators : Yes / No (If yes mention No)
)
17. Feed source
- a) Local /Branded//Both
 - b) If branded, Mention feed brand
 - c) FCR :
 - d) Cost of feed / Kg
(Rs): _____
 - e) Local factory name :
18. 1) Grow-out operations : a) Culture period _____ days
b) Harvest _____ days
c) Frequency of next harvest _____ days
d) Total harvests _____ no/crop
- 2) Best culture period for Grow out :
19. Water exchange : a) No of times per month
b) Percentage of exchange
20. Farm Management
- a) Tested water parameters :
 - b) Frequency of testing parameters :
 - c) Sampling frequency :
 - d) Recent disease problems :

21. Chemicals :
 a) Antibiotics :
 b) Sanitizers :
 c) Probiotics :
 d) Molt inducers :
 e) Others :
 f) Total cost on chemicals :
22. Crop Holiday : ----- days
23. Marketing :
 a) Agent /direct to company
 b) Head- on /.Head less

III. REASONS FOR CROP LOSS

24. A) Reasons for crop loss :
 i) seed Yes / No
 ii) Water quality Yes / No
 iii) feed quality Yes / No
 iv) climate Yes / No
 v) poor management Yes / No
 vi) poor technology Yes / No
 vii) Diseases Yes / No
- B) Loss period : From _____ to _____
- C) No. of crops taken so far ;
- D) No of successful crops ;
- E) No of failure crops ;

IV. TECHNICAL INFORMATION KNOWN

- 25 A) Have you undergone any training — Yes / No
- B) Do you want training if so what aspects you want Yes / No
- C) Do you aware of banned antibiotics ; Yes / No
- D) Do you like adopt new technology : Yes / No
- E) Do you know MPEDA guide lines ; Yes / No
- F) Do you know MPEDA code of conduct of practice : Yes / No
- G) Who is giving technical guidance :

26. Problems of shrimp farming :

1.

2.

3.

4.

41. Any Suggestions for Better Results :

1.

2.

3.

4.

Signature of the farmer

Signature of the scholar

Date :

Place:

State-wise Details of Shrimp Culture (as on 31 March 2002)

State	Shrimp	
	Area (ha)	Production (MT)
Andhra Pradesh	79 600	51 230
West Bengal	47 650	26 800
Orissa	8 120	8960
Kerala	1 4700	5540
Tamil Nadu	2 480	4710
Karnataka	3 080	3500
Goa	930	1200
Gujarat	540	680
Maharashtra	300	320
Total	1 57 400	1 02 940

Source : MPEDA (2003)

Order S.O. 528 (E) dated 17th May, 2002

S.O.528(E). - Whereas, in exercise of the powers conferred by section 6 of the Export (Quality Control and Inspection) Act, 1963 (22 of 1963), the Central Government has formulated certain proposals for amending the Order No. SO 729 (E) dated 21st August, 1995, of erstwhile Ministry of Commerce, Government of India, relating to Fresh, Frozen and Processed Fish and Fishery Products and in supersession of Ministry of Commerce and Industry Order No. S 729 (E) dated 17th August 2001 relating to Maximum Residual Limits (RRLs) for antibiotics, pesticides and heavy metals in fish and fishery products in the manner specified below for the development of export trade of India and has forwarded the same to Export Inspection Council as required by sub-rule(2) of rule 11 of the Export (Quality Control and Inspection) Rules, 1964;

Now, therefore, in pursuance of the said sub-rule, the Central Government hereby publishes the said draft proposals for the information of the public likely to be affected thereby;

Notice is hereby given that any person desiring to forward any objection or suggestion with respect to the said proposals may forward the same within thirty days from the date of publication of the said draft Order in the Official Gazette, to the Export Inspection Council, 3rd Floor, New Delhi YMCA Cultural Center Building, 1, Jai Singh Road, New Delhi - 110001.

PROPOSALS

In exercise of the powers conferred by section 6 of the Export (Quality Control and Inspection) Act, 1963 (22 of 1963), the Central Government after consulting Export Inspection Council, hereby makes the following amendments to the Order No. SO 729 (E) dated 21st August 1995 of erstwhile Ministry of Commerce, Government of India, relating to fresh, Frozen and Processed Fish and Fishery Products relating to Maximum Residual Limits (MRLs) for antibiotics, pesticides and heavy metals in fish and fishery products, which shall take effect on the date of the publications of the final proposals in the official Gazette namely :-

In said Order, in schedule - I, for clause (e), the following shall be substituted, namely :-

"(e) Maximum Residual Limits (MRLs) for pesticides, heavy metals and antibiotics and other pharmacologically active substances in fish and Fishery Products shall meet the requirements as given below :

Pesticides*

Pesticides	Max. permissible residual level in ppm
BHC	0.3
Aldrin	0.3
Dieldrin	0.3
Endrin	0.3
DDt	5.0

Heavy Metals*

Pesticides	Max. permissible residual level in ppm
Mercury	1.0
Cadmium	3.0
Arsenic	75
Lead	1.5
Tin	250
Nickel	80
Chromium	12

Antibiotics and other Pharmacologically Active Substances*

Pesticides	Max. permissible residual level in ppm
Tetracycline	0.1
Oxytetracycline	0.1
Trimethoprim	0.05
Oxolinic acid	0.3

(f) the use of any of the following antibiotics and other pharmacologically active substances shall be prohibited in the culture of or in any hatchery for producing the juveniles or larvae or nauplii of or in any unit manufacturing feed for or in any unit preprocessing or processing shrimp, prawns or any variety of fish intended for exports:

- (i) All Nitrofurans including
 - Furaltadone
 - Furazolidone

- Furylfuramide
- Nifuratel
- Nifuroxime
- Nifurprazine
- Nitrofurantoin
- Nitrofurazone
- (ii) Chloramphenicol
- (iii) Neomycin
- (iv) Nalidixic acid
- (v) Sulphamethoxazole
- (vi) Aristolochia spp and preparations thereof
- (vii) Chloroform
- (viii) Chlorpromazine
- (ix) Colchicine
- (x) Dapsone
- (xi) Dimetridazole
- (xii) Metronidazole
- (xiii) Ronidazole
- (xiv) Iprnidazole
- (xv) Other nitroimidazoles
- (xvi) Clenbuterol
- (xvii) Diethylstilbestrol (DES)
- (xviii) Sulfonamide drugs
(except approved Sulfadimeethoxine,
Sulfabromomethazine and Sulfaethoxypridazine)
- (xix) Fluoroquinolones
- (xx) Glycopeptides

*** Note:** However, for export to the European Union, the United States of America and Japan and other countries, the residual levels fixed by individual countries for specified products are to be complied with in respect of (e) and (f) above.

ORDER S.O. 1227 dated 23rd October, 2004

S.O. 1227 (E). - Whereas, for the development of the export trade of India, certain proposals for amending the order No. S.O. 729(E) dated 21st August 1995, of the erstwhile Ministry of Commerce, Government of India for prohibiting the use of substances having anabolic effect and unauthorised substances, veterinary drugs and contaminants, other substances and environmental contaminants in fish and fishery products were published in part II, sub-section (ii) of section 3 of the Gazette of India, Extraordinary, vide Order of the Government of India in the Ministry of Commerce and Industry, Department of Commerce, under S.O. 1035(E) dated the 9th September, 2003, as required under sub-rule (2) of rule 11 of the Export (Quality Control and Inspection) Rules, 1964 made under the Export (Quality Control and Inspection) Act, 1963;

And, whereas, the objections and suggestions were invited from all the persons likely to be affected thereby within a period of thirty days from the date of publication of the said Order in the Official Gazette;

And, whereas the objections and suggestions received from the public on the said proposals have been considered by the Central Government;

Now, therefore, in exercise of the powers conferred by clause (c) of section 6 of the Export (Quality Control and Inspection) Act, 1963 (22 of 1963), the Central Government, after consulting the Export Inspection Council, hereby makes the following amendments in the said Order No. S.O. 729(E) dated 21st August, 1995 relating to Fresh, Frozen and Processed Fish and Fishery Products which shall take effect on the date of its publication in the Official Gazette, namely: -

1. In the said Order, in Schedule I after clause (g), the following shall be inserted, namely:-

“(h) the use of any of the following substances having anabolic effect and unauthorised substances, veterinary drugs and contaminants and other substances and other environmental contaminants shall be prohibited in the culture of, or in any hatchery for producing the juveniles or larvae or nauplii of, or any unit manufacturing feed for, or in any unit pre-processing or processing, shrimps, prawns or any other variety of fish and fishery products, namely:-

- (i) substances having anabolic effect and unauthorised substances, namely:-
 - (a) stilbenes, stilbene derivatives and their salts and esters;
 - (b) steroids.
- (ii) Veterinary drugs and contaminants, namely:-
 - (a) antibacterial substances, including quinolones;
 - (b) anthelmintics.

(iii) Other substances and environmental contaminants
namely:-

- (a) organochlorone compounds including PcBs;
- (b) mycotoxins;
- (c) dyes.

Provided that the use of items at sl. No. (i)(b), (ii)(a) and (b) for therapeutic or zoo-technical purposes may be authorised by qualified Veterinary surgeons or Fishery Scientists."

F.No.6/2/2001-EI&EP

M.V.P.C. Sastry,
Joint Secretary.

Note : The principal order was published in the Gazette of India vide S.O. 729(E) dated 21st August, 1995 and subsequently amended vide S.O. 792(E) dated 17th August, 2001, S.O. 722(E) dated 10th July, 2002 and S.O. 464(E) dated 24th April, 2003.