

Ph. D Thesis

**STUDIES ON FISH WASTE MANAGEMENT PRACTICES IN
ARROOR SEAFOOD INDUSTRIAL BELT, KERALA AND
CONVERSION OF SEAFOOD WASTE INTO A
LIQUID PLANT GROWTH SUPPLEMENT**



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COCHIN UNIVERSITY OF SCIENCE AND TECHNOLOGY

November 2013

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ARROOR SEAFOOD INDUSTRIAL BELT, KERALA AND
CONVERSION OF SEAFOOD WASTE INTO A
LIQUID PLANT GROWTH SUPPLEMENT**

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In

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by

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Certificate

This is to certify that the Doctoral thesis entitled “**Studies on fish waste management practices in Aroor Seafood Industrial Belt, Kerala and conversion of seafood waste into a liquid plant growth supplement**”, is an authentic record of the original research work carried out by **Mr. Abhilash S.**, under my supervision and guidance at the School of Industrial Fisheries, Cochin University of Science & Technology, in partial fulfillment of the requirements for the degree of **Doctor of Philosophy** of Cochin University of Science & Technology and that no part thereof has been submitted before for any degree, diploma, associateship, fellowship or other similar recognition in any University or Institution.

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(Supervising Guide)

Declaration

I, **Abhilash S.**, do hereby declare that the thesis entitled “**Studies on fish waste management practices in Aroor Seafood Industrial Belt, Kerala and conversion of seafood waste into a liquid plant growth supplement**”, is an authentic record of work carried out by me under the supervision and guidance of **Prof (Dr) Saleena Mathew** at School of Industrial Fisheries, Cochin University of Science & Technology, in partial fulfilment of the requirements for the degree of Doctor of Philosophy of the Cochin University of Science & Technology, under the faculty of Marine Sciences and that no part thereof has been submitted before for any degree, diploma, associateship, fellowship or other similar recognition in any University or Institution.

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Abbreviations

APEDA	_	Agricultural and Processed Food Products Export Development Authority
ASIB	_	Aroor Seafood Industrial Belt
BHA	_	Butylated Hydroxyl Anisole
BIS	_	Beuro of Indian Standards
BOD	_	Biochemical Oxygen Demand
CCU	_	Chitin/Chitosan Units
COD	_	Chemical Oxygen Demand
COM	_	Compostable Organic Matter
CRD	_	Completely Randomized Design
DHA	_	Docosahexanoic Acid
DOSTE	_	Department of Science, Technology and the Environment
EPA	_	Eicosapentanoic Acid
ETP	_	Effluent Treatment Plant
EU	_	European Union
FAME	_	Fatty Acid Methyl Esters
FSN	_	Fish Soluble Nutrients
GDP	_	Gross Domestic Product
HACCP	_	Hazard Analysis Critical Control Point
ICP-AES	_	Inductively Coupled Plasma Atomic Emission Spectrometry
KSPCB	_	Kerala State Pollution Control Board
MAHACOT	_	The Maharashtra State Co-operative cotton Growers Marketing Federation Ltd.
MSW	_	Municipal Solid Waste
MUFA	_	Mono Unsaturated Fatty Acids
NHM	_	National Horticulture Mission
NPK	_	Nitrogen, Phosphorus and Potassium
NPOF	_	National Project on Organic Farming
PE	_	Polyethylene
PITC	_	Phenyl Isothiocyanate
PUFA	_	Polyunsaturated Fatty Acids
QMP	_	Quality Management Practices
RKVY	_	RashtriyaKrishiVikasYojana
RP - HPLC	_	Reverse Phase -High Performance Liquid chromatography
SFA	_	Saturated Fatty Acids
SSIS	_	Semi Structured Interview Schedule
ST	_	Surface Tension
TEA	_	Triethylamine
TPC	_	Total Processing Capacity
TQM	_	Total Quality Management
TSS	_	Total Suspended Solids
UNIDO	_	United Nations Industrial Development Organisation
USFDA	_	United States Food and Drugs Authority
WGP	_	Waste Generation Points
WHO	_	World Health Organisation

CHAPTER 1

GENERAL INTRODUCTION

Contents	1.1. Introduction
	1.2. Problem for the study
	1.3. Scope and significance
	1.4. Objectives of the study

1.1 Introduction

Waste management is an increasingly significant term often mentioned along with all important issues related to human intervention. As human settlement augmented, so do the issues associated with the waste generation. In the early ages of limited resource availability, less human density and availability of uninhabited land and other natural spaces, the problem of waste management was not that much a vital issue which needed to be tackled promptly. But as agricultural and industrial revolution accelerated the food and other goods production to surplus, it also simultaneously amplified the quantity and variety of waste generated. From household food waste, the spectrum of waste now extends to Municipal Solid Waste (MSW), industrial waste, biomedical waste, electronic waste and nuclear waste. The organic or biodegradable wastes, which form the lion share of the municipal solid waste category, exert the greatest challenge for management other than the hazardous waste. The high perishable nature of food and other bio-wastes makes them hard to handle and process before safe disposal.

The proliferation of various food industries throughout the world has further concentrated the problem. The seafood processing industry is no different in this aspect. Apart from contributing positively towards food security, livelihood and nutrition, the seafood sector also contributes immensely towards the organic waste pool of the world. The world's population is expected to increase by 36% during 2000 to 2030, from approximately 6.1 to 8.3 billion. It is also expected that the estimated total seafood demand will be 183 million tons by 2030 (Bastien, 2003). The accelerating increase in world population as well as the demand for seafood supply will subsequently result in further elevated seafood production and processing in every continent. This situation of elevated consumption shall also mean increased amount of waste generation in the seafood sector, which calls for some serious debates over the status of the waste management technologies, both existing and emerging in the future.

Pollution by waste, in all sense, is the most important negative externality generated by an industry. All the industries produce waste as a result of various manufacturing process followed by them. Unlike the chemical wastes, which has an immediate impact on the

environment whenever released above permitted limits, the organic wastes generated mainly by the food processing industries usually escapes unnoticed, until its impact on environment has reached uncontrollable proportions. According to United Nations Industrial Development Organization (UNIDO, 2010), the important environmental issues for the food industry include the following: Wastewater, which affects Biochemical Oxygen Demand (BOD); Total Suspended Solids (TSS); extreme nutrient loading specifically nitrogen and phosphorus compounds; pathogenic organisms and residual chlorine and pesticides. Solid Waste includes both organic and inorganic waste. Organic waste consists of the rinds, seeds, skin, and bones from processing operations and inorganic wastes normally include packaging materials like plastic, glass, and metal.

The seafood processing industry, one of the major food industries in the world also contributes to a large extent towards the negative externalities due to its peculiar nature of coastal specific location. The seafood is an important food source supplying around 13.8% and 16.5% of the animal protein consumed world-wide according to World Health Organization (WHO, 2013). World fish production touched 154 million metric ton in the year 2011 (FAO, 2012). The utilization pattern in 2011 showed that, 46.9 % of the fish for human consumption (134.8 million tonnes) was in live and fresh form. 53.1% of the world's fish production underwent different forms of processing for direct human consumption in frozen (29.3%), cured (9.8%) and prepared or preserved form (14%). Freezing was the most preferred method of processing fish, accounting for 55.2 % of total processed fish for human consumption.

India stands sixth in world inland and marine capture fisheries production (4.1 million tonnes) and second in aquaculture production with an average of 3.5 million tonnes in 2011 (FAO 2012). The seafood processing industry is one of the major players among the food processing sector of India, witnessing a considerable increase in infrastructure development in sync with the international standards over the years. At present there are about 369 fish processing units in India with a daily processing capacity of 10266 ton, out of which 257 are European Union (EU) approved. There are around 1050 registered exporters of various seafood items in the country among which 207 are exporters of chilled items, 149 exports dried seafood, 4 exports freeze dried products, 13 exports canned products, 555 deals with frozen products and 191 produces other seafood and allied products (MPEDA, 2013). The Indian seafood industry exported a total quantity of 813091 tonnes of seafood during the year 2010-11 worth 2856.92 million US\$. During this period, increase was noticed by 19.85% in quantity, 28.39% in rupee value, while the dollar value registered a 33.95% increase (MPEDA, 2013).

1.2 Problem for the study

One of the major challenges faced by the coastal communities especially in developing countries like India is the negative externalities exerted by the industries on their environment, affecting their livelihood. Even though the industries have created some positive impact in the society like increased job opportunities, infrastructure facilities etc., it is the negative ones that usually create an extended and deep impact on the local residents. All forms of processing

including value addition result in excessive quantities of waste. Depending upon the yield percentage of different seafood processing methods, a considerable quantity of fish waste is generated varying from as low as 3% to as high as 50%, till the finished product reaches the final customer. The seafood processing sector around the world thus generates considerable quantity of fish waste on a daily basis. Due to its high organic content, fish waste is often classified as a certified (prescribed) waste which is costly to dispose (Knuckey, *et al.*, 2004).

Indian environment is guarded by various environmental laws having strict instructions on solid and liquid waste management such as Environmental (Protection) Act, 1986, Water (Prevention & Control) Act, 1974 and The Municipal Solid Waste (Management and Handling) Rules, 2000. Despite this, the waste management scenario in India, especially of organic waste such as seafood processing waste, has been facing serious setbacks. The Indian seafood processing and allied industries like their counterparts elsewhere are mainly located in the coastal areas. The seafood processing and preprocessing units comply various quality standards like Codex standards, US FDA (United States Food and Drugs Authority) standards, EU (European Union) Norms, BIS (Bureau of Indian Standards) etc. at national and international levels. To achieve these standards several quality assurance programs are also developed and practiced. The HACCP (Hazard Analysis Critical Control Point) system of USA, the European Council directives, the QMP (Quality Management Practices) of Canada and TQM (Total Quality Management) of Japan are aimed to ensure safety and quality of fish and fishery products consumed in those countries. Even though the quality of the product is usually assured as per international standards, the same is not practiced in maintaining the quality of the environment into which the processing discards are discharged. The seafood solid waste contributes immensely towards the Compostable Organic Matter (COM) category of the Municipal Solid Waste (MSW) management system existing in the area. In most of the developing countries like India, municipal authorities lack the resources and trained staff, to provide their rapidly growing populations, with the necessary facilities and services for solid waste management to support an adequate quality of life (Bartone *et al.*, 1994).

1.3 Scope and significance

One of the most important requirements for a successful waste management programme is to have reliable data on waste generation rates, waste composition and management issues. Thus there is no alternative for a local analysis. The study aims at pioneering a quantitative evaluation of seafood waste generated at various levels of processing in the area. It also attempts to reveal a two way picture of the scenario by analysing the issues of waste management faced by the industry as well as the society in the context of environmental impact. Availability of quantitative data on seasonal and category wise generation of seafood waste, along with analysis of associated management issues could help in designing an efficient seafood waste management system, thereby reducing the ecological foot prints by the industry in the coastal zone. The utilization of the bulk quantity of seafood waste as foliar fertilizer opens a practical and productive solution for converting the liquid silage to plant foliage and vegetables,

thus ensuring increased organic productivity along with efficient waste management.

In the light of this, this study was materialized to analyze the management issues regarding the seafood processing waste generated, including its impact on the coastal community in one of the important seafood hubs of India, Aroor Seafood Industrial Belt (ASIB), Alappuzha District, Kerala. The area has witnessed serious pollution issues related to seafood waste, and seldom has any action been implemented by either the polluters or the preventers. Further this study is also intended to suggest a low cost, eco-friendly method for utilizing the bulk quantity of seafood solid waste generated in the area for the promotion of organic farming. The high nutritional value of seafood enables the subsequent offal to be considered as an excellent source for plant nutrition. The liquid silage, accepted worldwide as the cheapest and practical solution for rendering fish waste in bulk for production of livestock feed, is adopted in this study to develop foliar fertilizer formulations from various seafood waste. The effect of seafood foliar sprays is demonstrated by field studies on two plant varieties such as Okra and Amaranthus.

1.4 Objectives of the study

The major objectives of the study are:-

- To determine the current status of seafood waste generation, management and utilisation in Aroor Seafood Industrial Belt (ASIB), Alappuzha District, Kerala.
- To assess the extent of impact generated by the current waste management practices over the coastal community in the area.
- To analyse the nutritional quality of fresh seafood waste available in the area.
- To develop foliar fertilizer formulations from seafood waste using organic acid ensilation technique.
- To study the effect of seafood foliar sprays on plant varieties such as Okra and Amaranthus.



CHAPTER 2

SEAFOOD WASTE GENERATION AND MANAGEMENT: A REVIEW

Contents	2.1 Introduction
	2.2 Pollution potential of seafood processing wastes
	2.3 Status of seafood waste management methods adopted world wide
	2.4 Role of waste minimisation and cost effectiveness in sustainable seafood waste management

2.1 Introduction

The process of seafood waste generation starts right when the fish is landed on the deck of the boat after capture. According to FAO (2010), about 7 million tonnes of fish harvested around the world was discarded back to sea, contributing significantly towards the oceanic pollution. After surpassing the onboard sorting, further loss is registered when the seafood material reaches the shore. The percentage of loss increases according to the non-availability of proper post-harvest seafood handling, storage and transportation facilities. Akande and Diei-Ouadi (2010) projected the post-harvest fish losses in developing countries are up to 40% of total landing, due to improper handling, processing and marketing infrastructure. The seafood waste generation pattern and yield also depends upon factors like the type of raw material used, method of processing and packaging practiced etc. In Table 2.1, an input of raw material and output of liquid and solid wastes from various processing operations are depicted.

2.1.1 Seafood solid waste generation

Seafood could be widely classified on a commercial trade basis into three major varieties such as the shell fish, fin fish and cephalopods. The shellfish group consists of the crustaceans such as shrimps, lobsters, crabs and molluscs like clams, oysters and mussels. The fin fish consists of all the major varieties of commercially important fishes such as sardine, mackerel, tuna etc. The cephalopods cover the seafood varieties such as the cuttlefish, squid and octopus.

Table 2.1: Input of raw material and output of liquid and solid wastes from various fish processing operations

	Outputs/1000 kg	
	Liquid waste (m ³)	Solid waste (kg)
White fish filleting	5-11	Skin: 40-50
Oily fish filleting	5-8	400-450
Canning	15	Heads/entrails: 250
Frozen fish thawing	5	-
De-icing and washing	1	0-20
Grading	0.3-0.4	0-20
Scaling of white fish	10-15	Scales: 20-40
De-heading of white fish	1	Heads and debris: 270-320
Filleting of de-headed white fish	1-3	Frames and off cuts: 200-300
Filleting of un-gutted oily fish	1-2	Entrails, tails, heads and
Skinning white fish	0.2-0.6	Skin: 40
Skinning oily fish	0.2-0.9	Skin: 40
Trimming and cutting white fish	0.1	Bones and cut-off: 240-340
Unloading fish for canning	2.0-5.0	-
Grading of fish	0.2	0.30
Knobbing and packing in cans	0.2-0.9	Heads and entrails: 150
Skinning of knobbed fish	17.0	Skin: 55
Precooking of fish to be canned	0.07-0.27	Inedible parts: 150
Draining of cans containing	0.1-0.2	-
Washing of cans	0.04	-
Sterilization of cans	3.0-7.0	-

(Adapted from Arvanitoyannis and Kassaveti, 2008)

Processing of fish for fillets generates a bulk amount of waste up to 60% of the raw material and most of which is discarded (Adeyemo, 2013). Fish head waste constitutes around 20% of the fresh water fish biomass (Bhaskar, 2008). About 70% of tuna is lost as waste during various processing stages of canning (Herpandi *et al.*, 2012). The production of waste varies from species to species. Fatty fishes depending on their seasonal fat content, varying from 2% to 25%, exert significant impact on the pollution load. It has been reported that due to the presence of significantly high composition of exoskeleton and other non-edible internal organs, the shell fishes produces a large quantity of waste.

Archer (2004) reported production of around 68% of waste from crabs, 65% from shrimps, 56% from lobsters and 80% from molluscs on the basis of average percentage of live weight. The head, shell and tail portions of shrimp account for about 50% of the volume of raw materials (Islam *et al.*, 2004). The shrimp waste consists of 34-45% head and 10-15% shell (Ramayadevi *et al.*, 2012). Crabmeat is available as fresh, pasteurized or in canned form. The snow crab processing waste has been reported to be 25% of the total weight (Stewart and Noyes-Hull, 2010). Oyster shells vary considerably in thickness according to their origin and as a result, the percentage yield of meat also varies. The yield of meat from oysters normally ranges from 6 to 18 %, and from Pacific oysters 5 to 14 % (Stroud, 2001). Azanza *et al.* (2003) reported a final meat yield ranging from 33.04 to 34.38% from mussel processing.

Dabrowski *et al.* (1970) reported 65.7% average yield of edible flesh for cuttlefish. The yield of squid species is reported to be higher than fin fishes (60 to 70%) by Stroud (2001). Aberdeen (1989) reported that the edible meat yield of octopus species is about 78-80%.

2.1.2 Seafood liquid waste generation

Wastewater from seafood processing facilities is generated as a result of a variety of processes such as raw material unloading, cleaning, equipment washing, disinfection and facility sanitation (Morry *et al.*, 2003). Fish factories in Japan have calculated water consumption to the tune of 18–60 L/kg finished product for various types of processing plants (Islam *et al.*, 2004). According to a study conducted by Department of Science, Technology and the Environment (DOSTE), Vietnam (2003), normal cleaning of table and 4-5 m² of factory floor consumed about 2-3 m³/hour of fresh water. Large quantities of water (18–27 m³) per ton raw material are usually used in peeling machines (Nordic Council, 1997). The process of cooking squid mantle muscle generates a considerable amount of effluent with high organic load, resulting in high treatment cost and the effluent can contaminate the adjacent areas if discarded into the sea (Zaidy *et al.*, 2010). Typical effluent flow rates of the squid processing facilities were estimated to be 15,000 to 20,000 gpd (Gallons per day) by Park *et al.* (2001).

2.2 Pollution potential of seafood processing wastes

The health risk associated with organic wastes is the presence of high concentrations of pathogenic organisms and their potential to spread diseases (WHO, 2006). The solid and liquid wastes generated from various seafood processing activities are generally classified under the organic or biodegradable waste category. Seafood solid waste dominates this category.

The solid wastes are high in organic components such as Nitrogen, Phosphorus, and Potassium which are useful in fertilizing the soil when applied in limited concentrations. When handled as waste, and dumped to an open environment without following a safe and environmentally friendly protocol, the highly perishable solid wastes such as seafood offal could create serious environmental issues. The solid waste exposed to external elements undergoes decomposition aerobically and anaerobically as a result of the action of different microorganisms that feed and proliferate on the organic matter in the seafood waste (De Bertoldi *et al.*, 1983). The risk to the environment is brought by the proliferation of pathogenic microorganisms and the leachate generated by the deteriorating organic waste landfills. Landfill leachate is generated by surplus rainwater passing through the landfill (Kjeldsen *et al.*, 2002).

Other than the pollution caused by the leachate from solid wastes, the seafood processing activities also generates a considerable amount of liquid effluent, which when released to the environment, creates even more dangerous levels of water pollution. The seafood processing plant effluent contains significant quantity of organic matter in the form of small particles of flesh, breading, soluble proteins, and carbohydrates. According to United Nations Industrial Development Organisation (UNIDO), the key environmental issues of concern regarding the waste water generation from food processing industries are increased levels of biochemical oxygen demand (BOD); chemical oxygen demand (COD), total suspended solids (TSS); nutrient loading, namely nitrogen and phosphorus compounds; pathogenic organisms and residual chlorine and pesticide levels. These parameters indicate the strength of the effluent and have been standardized worldwide by various environmental regulatory agencies with specific limits in order to permit safe discharge to the environment.

A comparison of data regarding the actual strength and quantity of seafood processing wastes with the permissible limits of discharge reveals the magnitude of the problem of them getting into the open ecosystems in an unregulated manner. The problem occurs when the quantity of the organic matter discharged is too large. The capacity of the ecosystem to recycle the organic matter is then interrupted which results in significant changes. Other than reducing the level of oxygen dissolved in water, the biodegradation of this organic matter initiates various changes in the chemistry of the water body like increase in the ammonia, excess concentration of nitrogen and phosphorus based nutrients, pH variation, increased turbidity of water etc. (Nadia *et al.*, 2004). This also results in an increase in phytoplankton biomass and decrease in species diversity with benthic and fish communities on a large scale in long term (Bonsdorff *et al.*, 1997). Excess nutrients from organic matter decay increase plant growth which together with oxygen depletion could lead to alterations in ecosystem structure and eutrophication. Eutrophication, which promotes proliferation of macro algae and filamentous algae, creates havoc affecting benthic fauna, nursery and feeding of fish, recreational uses and tourism (Riegman, 1995).

Effluents from shrimp canning have been reported to have concentrations of BOD, COD, Oil and Fat, Total Nitrogen (TN) and NH₃-N of 1081, 2296, 258, 196, and 802 mg/l respectively with corresponding net waste loads of 46, 109, 11, 7.6, and 37.7 g/kg (Carawan, 1991).

Park *et al.* (2001) reported that processing of squid and several types of finfish creates high levels of BOD ranging from 1000–5000 mg/l. Squid ink released into the waste watercourse during processing contains high concentrations of organic matter, as well as soluble proteins which contribute significantly to excessive BOD load (Shirai *et al.*, 1997). Table 2.2 depicts the Government of India standards for effluent discharge for canning and food processing industries.

Table 2.2 Effluent discharge standards for canning and food processing industries

Parameter	Unit	Maximum permissible limit	
		Land/Under ground	Surface water courses
Total coliforms	MPN per 100 ml	-	<400
E. Coli	MPN per 100 ml	<1000	<200
Free Chlorine	mg/l	-	0.5
Total Suspended Solids (TSS)	mg/l	45	35
Reactive Phosphorus	mg/l	10	1
Temperature	°C	40	NA
pH	-	5 – 9	NA
Chemical Oxygen Demand (COD)	mg/l	120	NA
Biochemical Oxygen Demand	mg/l	40	NA
Chloride	mg/l	750	NA
Sulphate	mg/l	750	NA
Sulphide	mg/l	0.002	NA
Ammonia Nitrogen	mg/l	1	NA
Nitrate as N	mg/l	10	NA
Total Kjeldahl Nitrogen (TKN)	mg/l	25	NA
Nitrite as N	mg/l	1	NA

Source: Environment Protection (Standards for effluent discharge) Regulations 2003, Government of India.

2.3 Status of seafood waste management methods adopted world wide

The seafood wastes need to be treated by various means in order to render them fit for safe disposal. Both solid and liquid wastes are required to be handled and managed according to their physical state, composition and pollution potential.

2.3.1 Seafood solid waste management

Seafood solid wastes are generated in huge quantities in the processing zones which makes them impossible to be utilized entirely for recycling and reuse in order to produce by-products. In this scenario the solid wastes are usually treated as other organic wastes and are included in the Municipal Solid Waste (MSW) category to be handled by the government authorities. This further complicates the problem as the higher moisture content and organic components render the seafood wastes susceptible to a higher rate of deterioration, which makes them difficult for handling (Knuckey, *et al.*, 2004). This particular nature of the seafood wastes mostly affects the developing countries which lack sufficient machinery for waste management as observed by Wang *et al.* (2011). Developing nations generates less quantity of waste per capita, with a higher amount of organic (biodegradable) material in the municipal solid waste, which is estimated to be 50% of total waste when measured by weight (UNEP, 2005).

Most of the MSW in developing countries is dumped on land in a more or less uncontrolled manner due to nonconformity of procedure for setting, design, and operation of new landfills and lack of recommendations for upgrading existing open dumps (Da Zhu *et al.*, 2008). The disposal methods in some of the countries indicate the share of open dumping to be 90% in India, 85% in Sri Lanka, 65% in Thailand and 50% in China (Anubhav *et al.*, 2011). Most of the current landfills are covered in the dumpsite by soil without any proper technical input and disregarding the importance of treatment of the emerging emissions to water, air and soil (Visvanathan and Trankler, 2004). Due to the peculiar characteristics of the seafood solid waste as discussed above, landfilling and incineration are least preferred for disposing them. The best option for managing seafood waste is to channel it for by-product recovery because of its high nutritional quality. When the seafood waste quality is beyond the condition of being utilized for other by-product production, the most basic and last resort of processing seafood solid waste is composting (Schaub *et al.*, 1996).

2.3.1.1 By-product recovery as a tool for seafood solid waste management

An important waste reduction strategy for the industry is the recovery of marketable by-products from seafood wastes. The utilization of seafood solid waste could be used by the industry as a tool for cleaner production as this creates additional revenues as well as reduction in waste disposal expenses (Arvanitoyannis and Kassaveti, 2008).

The most popular methods for utilization of seafood waste are manufacture of fishmeal/oil, production of seafood silage and the use of fish waste in the manufacturing of organic fertilizer (Gill, 2000). Fish waste rich in vital nutrients opens up an array of by-product options for recovering such as fish meal, fish oil, bio-fuel, fish silage, organic fertilizer, fish protein

hydrolysate, enzymes, collagen and bioactive peptides. Extensive research have been carried out to develop methods to convert fish waste into useful products (Kristinsson and Rasco, 2000; Larsen *et al.*, 2000; Guerard *et al.*, 2001; Coello *et al.*, 2002). The shellfish waste consists of the shell and head materials of crustaceans from both capture fisheries and aquaculture. Conventionally crustacean waste has been disposed through incineration, landfilling and ocean dumping (Peberdy, 1999). With a history of positive and negative handling of solid and liquid waste, the seafood industry is now trying to answer the issues of cost involved in transporting of waste, environmental pollution and wasting 70 to 80% of the dry weight of the catch through finding alternative methods of waste utilisation (Vyas and Deshpande, 1991). Table 2.3 depicts seafood solid waste utilization options studied by various researchers.

Table 2.3 Seafood solid waste utilization options

By-product	Reference
Finfish waste	
1. Fishmeal	Ponnusamy <i>et al.</i> , 2012; Hall, 2010.
2. Fish oil	Rubio-Rodríguez <i>et al.</i> , 2012; Khoddami <i>et al.</i> , 2012.
3. Bio-fuel	Yahyaee <i>et al.</i> , 2013; Cherng-Yuan and Rong-Ji, 2009.
4. Fish silage	Wicki <i>et al.</i> , 2012; Zynudheen <i>et al.</i> , 2008.
5. Organic fertilizer	Kim, 2011; El-Tarabily <i>et al.</i> , 2003.
6. Fish protein hydrolysate	Benhabiles <i>et al.</i> , 2012; Bhaskar <i>et al.</i> , 2008.
7. Enzymes	Temiz <i>et al.</i> , 2013; Tavares <i>et al.</i> , 1997.
8. Collagen & Gelatin	Zhang and Jin, 2011; Songchotikunpan <i>et al.</i> , 2008
9. Bioactive peptides	Wang <i>et al.</i> , 2013; Picot <i>et al.</i> , 2006.
Shellfish waste	
1. Shell waste derivatives	Vázquez <i>et al.</i> , 2013; Mahmoud <i>et al.</i> , 2007.
2. Pigments	Amir <i>et al.</i> , 2012; Chakrabarti, 2002.
3. Shell fish hydrolysates	Dey and Dora, 2011; Bueno-Solano <i>et al.</i> , 2009.
Cephalopod waste	
1. Collagen	Kreigand Mohseni, 2012; Uriarte-Montoya <i>et al.</i> , 2010.
2. Cephalopod hydrolysates	Lin <i>et al.</i> , 2012; Wang <i>et al.</i> , 2011.
3. Cephalopod Oil	Uddin <i>et al.</i> , 2010; Yoshida and Tavakoli, 2006.
4. Cephalopod ink	Nan-Nan and He-Sheng, 2011; Naraoka <i>et al.</i> , 2000.

2.3.2 Seafood liquid waste management

Water is the most important component required for hygienically processing seafood for producing a superior quality end product. Water is mostly used in food processing as a component, medium and sanitation aid (Kirby *et al.*, 2003). Use of water thus generates a considerable quantity of effluent waste especially in sea food processing which has been blamed of generating highly polluted wastewaters (Veiga *et al.*, 1994) and about half of the organic industrial pollution by the food industries (Maxime *et al.*, 2000). Advancement in processing techniques together with the expansion plans in production, have led to remarkable increases in the volume and strength of wastewater discharged by fish processing industries (Park *et al.*, 2001).

A standard process for treating high organic load effluents like from seafood processing consists of primary and secondary treatment stages. Table 2.4 depicts the criteria followed for deciding the magnitude of effluent treatment adopted in a seafood processing unit.

Table 2.4 Identification criteria for wastewater treatment options in seafood processing:

1	Levels of settleable and suspended solids, oil & grease.	<ul style="list-style-type: none"> Pre-screening, grit removal & Flotation unit.
2	Levels of BOD/COD	<ul style="list-style-type: none"> Typically < or > 1000 mg/l (Aerobic vs. Anaerobic)
3	Levels of organic nitrogen & ammonia	<ul style="list-style-type: none"> Extended aeration for nitrification
4	Requirement for de-nitrification	<ul style="list-style-type: none"> Incorporation of anoxic zone
5	Total Phosphate content	<ul style="list-style-type: none"> Biological uptake/precipitation
6	Disinfection requirements	<ul style="list-style-type: none"> Oxidizing agent (Cl₂/O₃)
7	Sludge treatment requirements	<ul style="list-style-type: none"> Sludge thickening, dewatering

(Adapted from Duangpaseuth *et al.*, 2009).

The primary treatment consists of employing physical treatment methods such as screening, sedimentation etc. and secondary treatment involves chemical and biological methods. Seafood processing facilities mainly apply an amalgamation of primary and secondary wastewater treatments, depending on the strength of the effluent stream. (Duangpaseuth *et al.*, 2009). A detailed classification of seafood effluent treatment methods currently followed worldwide is given in Figure 2.1.

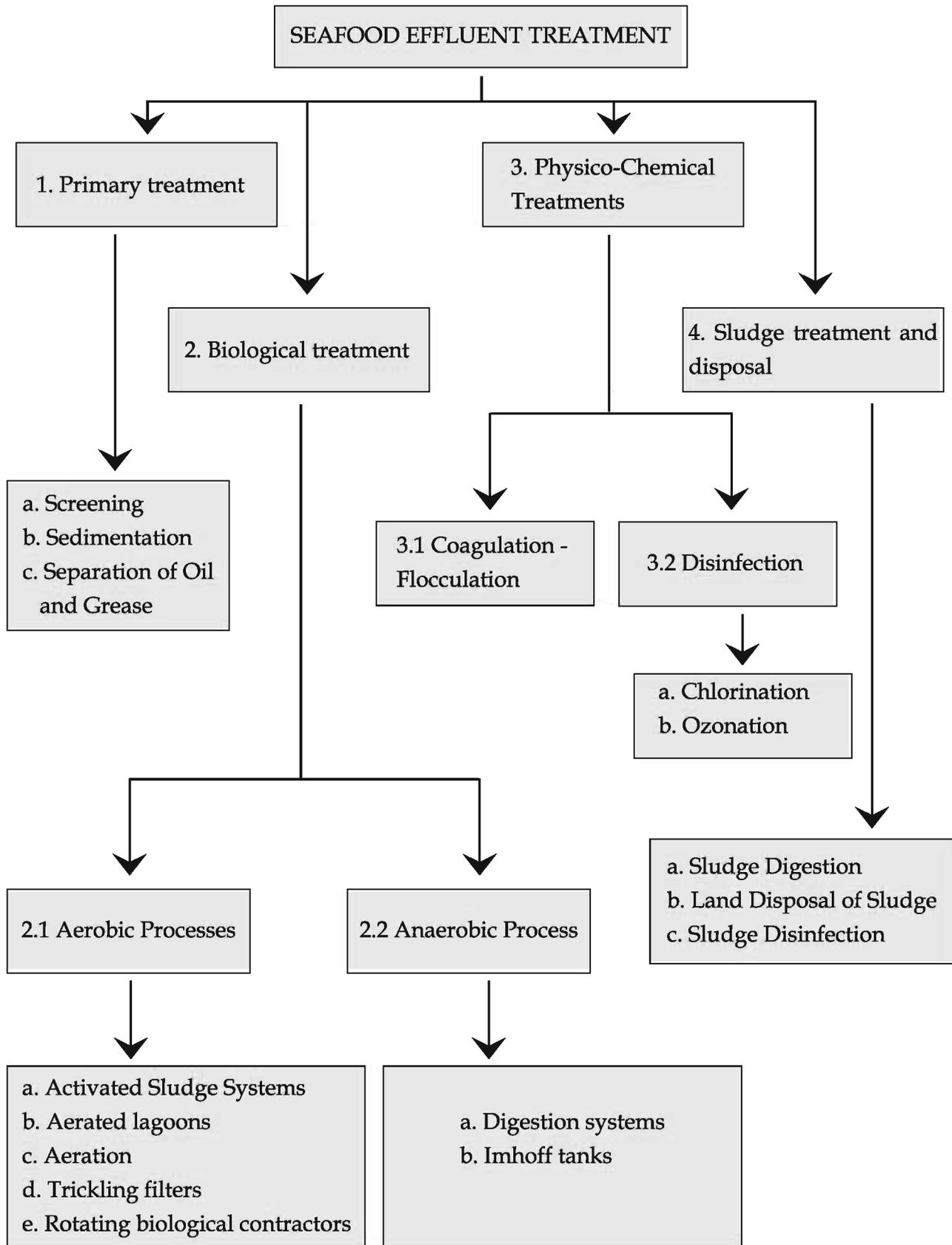


Figure 2.1 Seafood effluent treatment methods classification (Adapted from, Genovese and González, 1998)

2.3.2.1 By-product recovery as a tool for seafood liquid waste management

Like the solid wastes generated during the seafood processing, the liquid waste also opens up possible resource recovery options as it carries valuable organic components of the raw material subjected for processing such as protein, enzymes, pigments, aroma components and oil. Even though some authors have observed that reuse and recycling of waste materials are on the whole difficult to apply in food industries (McDonald *et al.*, 1999). Asbjorn (2004) suggested that better returns can be obtained by making products for industrial use and human consumption such as biopolymers, enzymes and bioactive peptides. Table 2.4 depicts seafood liquid waste utilization options proposed by various researchers.

Table 2.4 Seafood liquid waste utilization options

By product	References
Protein recovery	Pérez-Gálvez <i>et al.</i> , 2011; Afonso and Bórquez, 2002a.
Oil recovery	Sala, 2012; Chang-Wei Hsieh <i>et al.</i> , 2005.
Aroma compounds	Walhaa <i>et al.</i> , 2011; Cros <i>et al.</i> , 2004.
Enzymes & pigments	Stepnowski <i>et al.</i> , 2004; DeWittand Morrissey <i>et al.</i> , 2002.

2.4 Role of waste minimisation and cost effectiveness in sustainable seafood waste management

The increasing awareness on the impact created by food wastes on the environment has urged the authorities to impose more and more stringent regulations on their handling and disposal in recent times. This has considerably increased the cost incurred by the manufacturing industries on waste containment, treatment and transport. With the ever increasing demand on food items especially like seafood, the quantity of raw material processed seems to be increasing by time. In this scenario, efforts have to be made towards waste minimization by introducing efficient processing protocols and low cost waste treatment technologies. In the case of seafood industry, efficient water management, introduction of new high yield processing technologies and efficient recycling are the major tools for sustainable waste management.

United Nations Industrial Development Organization (UNIDO, 2010) has predicted that pre-treatment and water conservation will be chief targets for pollution prevention in the food-processing industry, aiming to minimize raw materials loss to the processing waste water. Casania *et al.* (2006), observed that the increasing costs of water and water discharge along with reduced water availability and environmental problems have increased the interest on the reuse of water in food production and processing, both from an economical and sustainability point of view. They also indicated that the most significant water savings have been achieved by reducing uncontrolled use and improving layout design, planning, and control in both production and cleaning procedures. The most significant EU document (Directive 98/83/EC, 1998) dealing with water quality and its use in the food industry consents to the use of water of different qualities to drinking water in food production, after careful assessment and authorization

by the proficient national establishments. Water recycle has become important to seafood producers due to rising utility costs, limited water resources, and pollution problems associated with disposal (Afonso and Borquez, 2002b).

After minimising the quantity of solid and liquid wastes generated through improved processing and water management systems, another important area to be concentrated for achieving the goal of sustainability in seafood waste management is the proper treatment of waste generated. The solid waste handling and management cost could be considerably reduced by adopting suitable by-product valorisation technologies as discussed in previous sections. In the case of liquid waste, the magnitude of treatment is decided by the pollution strength of the effluent generated. Tchoukanova *et al.* (2004) observed that the cleaning procedures in seafood processing consumes about 25 to 40% of the total volume of potable water used during the production and thus recommends application of dry cleaning prior to washing i.e. removing the solid particles without giving a chance for getting contacted with water and thereby reducing the organic load on the effluent generated. Some of the studies from developing countries (Sasidharan and Mathew, 2011) show that the cost involved in effluent treatment seriously affects the adaptation and extend of their operation by the processing firms. They observed that under-operation of effluent treatment plants (ETP) by some of the processing units were attributed to the high constructional and operational costs of the units. This kind of scenarios point to the need for the popularization of cost efficient ETP technologies.

Management of biodegradable waste is equally an increasing concern among the developing and industrial nations. Seafood waste, like all other organic processing wastes is difficult to handle, transport and to treat prior to disposal, and on the other hand poses serious impact to the immediate environment if left out without any treatment. Studies show that a considerably huge quantity of seafood solid waste is generated as a result of the inefficient harvesting, post-harvest handling, storage and transport facilities worldwide. The lower product yield of existing processing method preferences also adds up to the extensive generation of seafood solid waste. On the other hand indiscriminate usage of potable water in various stages of seafood raw material handling right from onboard the fishing vessel to packaging in factory is generating enormous quantity of seafood effluent adding immensely towards the waste pool. Significant variation in waste yield and sources such as fishes, shellfishes and cephalopods also render the seafood waste difficult to be standardized under a common waste management protocol.

Existing solid waste management systems such as land filling, composting and incineration does not hold much promise towards the environment safety in the future considering the huge quantity of seafood solid waste generated on a daily basis. Extensive research have been made on valuable by-product development possibilities from different varieties of seafood waste and most of them are unfortunately on lab scale till date. Efforts have to be made on scaling up the processes on an industrial scale with emphasis on the cost and energy efficiency. By-product recovery could be incorporated into the solid waste management scheme of an industrial seafood unit, thus providing additional revenue, reduction in waste management cost and lesser load over the environment.

The seafood liquid waste management is currently more organized in the sense of the existence of a widely accepted and almost standardized treatment protocols such as mechanical filtering, sedimentation, biological and chemical treatments prior to disposal. The concept of waste minimization is gaining momentum in this sector by introducing water management protocols, high yield processing technologies and machineries, by-product recovery from effluents and improved solid waste handling. Reduction in quantity and strength of seafood effluents along with introduction of cost and energy efficient technologies like product recovery, membrane filtration etc, could considerably change the current seafood waste management scenario into the emerging sustainability concept.



CHAPTER 3

STATUS OF SEAFOOD WASTE MANAGEMENT IN AROOR SEAFOOD INDUSTRIAL BELT, ALAPPUZHA, KERALA

Contents	3.1 Introduction
	3.2 Background of the study
	3.3 Methodology
	3.4 Results and Discussion
	3.5 Impact of seafood waste on coastal livelihood: A case study
	3.6 Conclusion

3.1 Introduction

Seafood has been an integral part of the diet of the people of Kerala for centuries especially along the coastal belt that stretches around 600 Km. The demand and supply of seafood in Kerala has been constantly on a high note which is materialised through the rich resources in the fisheries sector of the state. The state stands second in total marine fish production (SPB, 2012) in India and third in quantity of seafood export from the country (KMFS, 2010). Table 3.1 depicts a general fisheries profile of Kerala.

Table 3.1: Fisheries Profile of Kerala

Coastline of Kerala	590 Km.
Marine fishing villages	222
Inland fishing villages	113
Fishing harbours	10
Fish landing centres	61
Wholesale fish markets	185

Retail fish markets	2518
EU approved processing plants	62
Prawn filtration fields	6,129 hector
Public sector brackish water farms	2,873 hector
Fishing crafts	21746
Marine Fisherman Population	879800
Marine fish production	570013 MT
Inland fish production	116836 MT
Export through the ports of Kerala	
Quantity in M. Tonnes	107293
Value in ₹ Cr.	1670
Percentage share of Kerala in terms of Quantity in export	16
Percentage share of Kerala in terms of Value in export	17

Source: KMFS, 2010, Directorate of Economics & Statistics, Govt. of Kerala

The lion share of the seafood export from Kerala is through the Cochin port, in Ernakulam District which has resulted in concentration of seafood and allied industries in and around the area. The adjacent Aroor Municipality in Alappuzha District have jointly become a major seafood processing hub in Kerala and in India as a whole, highlighted by the highest concentration of processing and pre-processing units in the country. This elevated concentration of seafood industrial activities has also resulted in major issues related to waste management having serious environmental and social impacts in the area.

3.2 Background of the study

The Aroor Seafood Industrial Belt (ASIB) as evident from the media reports (Plate 3.1) has been continuously making news for being an area of severe environmental crisis affecting the day to day survival of the local community. Irrespective of the temporary corrective actions taken by the regulatory authorities, the issues seems too persistent for a long period of time. The major responsibility for the environmental degradation in the zone has been pointed towards the seafood processing industry which is one of the major industrial activities in the area. The

major responsibility for the environmental degradation in the zone has been pointed at the seafood processing industry which is one of the major industrial activities in the area. The media and the authorities has been accusing the industry for irresponsibly discharging solid and liquid waste to the adjacent water bodies without performing mandatory waste treatment procedures. The presence of natural interconnected water canals in the area, on which the local population depends for water and other livelihood requirements, also magnified the effects of pollution resulting in serious social resistance.

The connection of the water canals with the Vembanad Lake, one of the ecologically significant water bodies of the country, also makes the issues environmentally momentous. The Vembanad Lake, one of the major estuaries in India is situated between latitudes 9°28' and 10°10' N and longitudes 76°13' and 76°31' E in southern Kerala with a length of 90 km extending from Alleppey to Azheecode with water spread area of 300 sq. km. Various studies have indicated the role of industrial activities like seafood processing in a variety of pollution issues in the area. Laxmilatha and Appukuttan (2002) in their study observed that pollution caused by retting of husks, effluent discharged from industries and environmental hazards caused by the effluent released from shrimp peeling sheds to the Vembanad Lake as the main reason for the shrivel in clam production. Abraham and Ravindran (2009) in their study on the sustainable duck rearing in Kerala has indicated that feeding of fresh prawn waste generated in the Aroor area to ducks could solve the crisis of water and air pollution, widespread in the area for a long time due to disposal of prawn waste into the water bodies. Mathew et al. (1989) also pointed out industrial pollution, husk retting and pesticide discharge are the major reasons for fluctuations of the physiochemical parameters of the Vembanad Lake, indicating the persistence of the problem for a period of time. The existing and limited studies on the issues lack a clear picture comprising the giving and receiving ends of the pollution spectrum. The studies largely concentrated on the receiving end, projecting the changes in physiochemical parameters of the polluted water bodies. The seafood processing is a complex and energy/resource consuming activity which needs to be closely evaluated for the complete analysis of the situation.

3.3 Methodology

3.3.1 Study area profile

The area selected for the study, Aroor Seafood Industrial Belt (ASIB), is located in the Alappuzha district, Kerala, India (Fig. 3.1). ASIB extends to a length of 7.6 Km covering Aroor, Chandiroor, Eramalloor, Ezhupunna, Kuthiyathode and Thuravoor Panchayats (Local governing bodies). According to 2001 India Census, Aroor, which is a census town, is having a population of 35,281. Males constitute 49% of the population and females 51%. Chandiroor Village is situated along NH-47, 24 km south of Cochin and it consists of around 3450 houses with a total population of 11,500, with majority of the people hailing from a poor socio-economic stratum. The Eramalloor village has a population of 28223 with 14187 males and 14036 females. The total population of Ezhupunna village is around 27206 with 13214 males and 13992

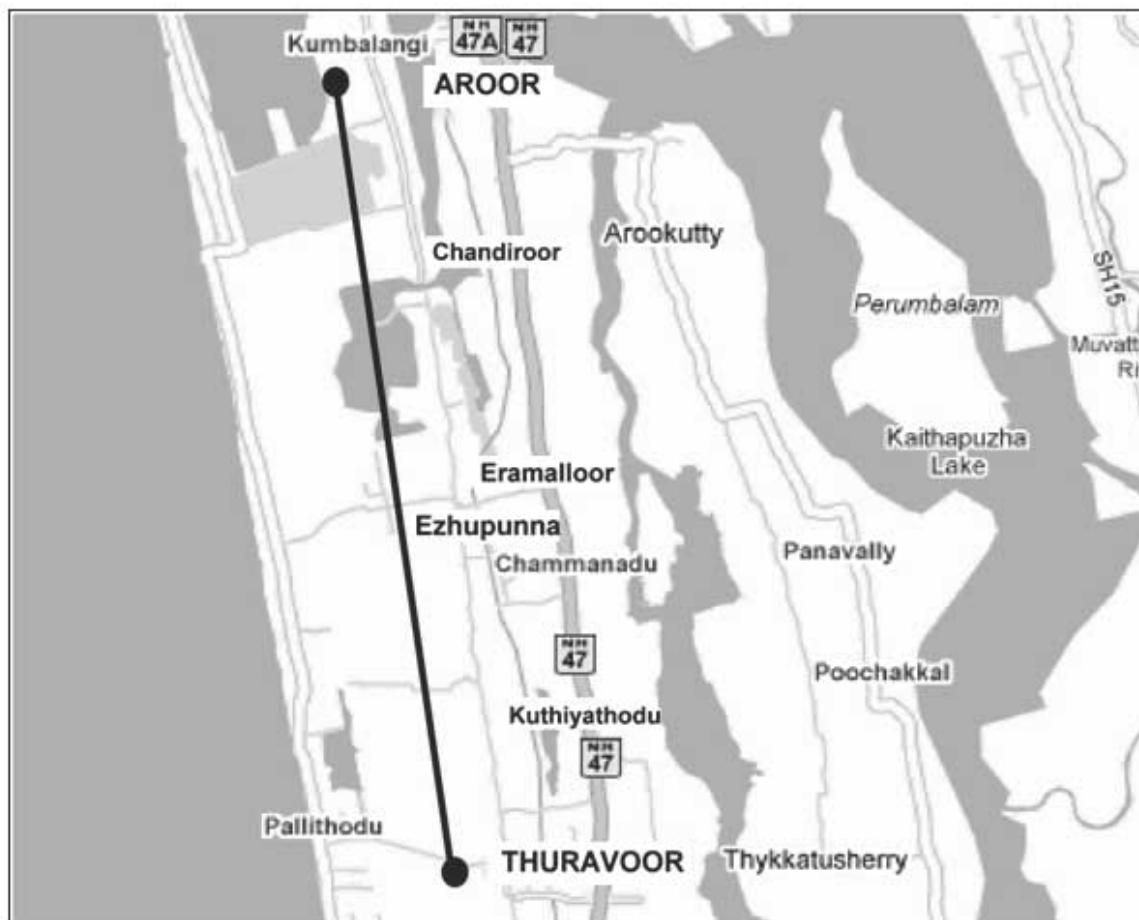


Figure 3.1 : Map showing the stretch of Aroor Seafood Industrial Belt (Source: Google maps)

females. The Kuthiathode village harbors a population of 22,880 with 11,098 males and 11,782 females and the Thuravoor village has a population of 27838 with 13417 males and 14421 females. All the villages including Aroor come under the Aroor Assembly Constituency. Fishing and allied industries are their major source of livelihood. The place is endowed with a network of canals which are invariably connected with either the Arabian Sea on the west or the Vembanad back waters on the east side. Income generating activities other than seafood in the area are coir making from coconut husk, livestock rearing, agriculture and fishing. The proximity of ASIB to Cochin, the commercial capital of Kerala, has considerably helped in elevating its status both industrially and economically, by expanding its reach to two important fishing harbours, Thoppumpady and Munambam, as well as the international seaport and airport facilities available at Cochin.

3.3.2 Survey methodology

A ban on mechanized fishing, especially trawling is exerted by the Government of Kerala in the coastal waters of the state for a period of 45 days, during the month of June and July every year. As the trawl fishery is the major shareholder in the marine fish landings of Kerala, a significant difference in the quantity of raw material handled by the seafood processing sector in the study area during this period is visible.

Keeping this in mind, the study period was divided into: -

- (a) Pre trawl ban period from January to May
- (b) Trawl ban period from June to July
- (c) Post trawl ban period from August to December.

3.3.3 Data acquisition:

3.3.3.1 Primary Data:

The primary data regarding the waste management status and issues were collected through census method (Fig. 3.2) using Semi Structured Interview Schedule (SSIS) questionnaires (Annexure 3.1 – 3.5) specifically prepared for the survey units such as seafood processing units, pre-processing units, fish markets (Plate 3.2 a-c) and shell waste utilizing units. Similar approach was practiced for gaining information from the local residents in the area regarding the impact of current waste management system (Annexure 3.6). Around 300 families comprising of around 1500 individuals residing in the proximity of the Chandiroor canal, the pollution hot spot of the zone, were identified for studying the social impact of the existing seafood waste management system. Thirty percentage of the individuals, randomly selected, was subjected to face to face interview. Data regarding the social issues were also collected from Key Informants in the area such as local leaders, health workers etc.

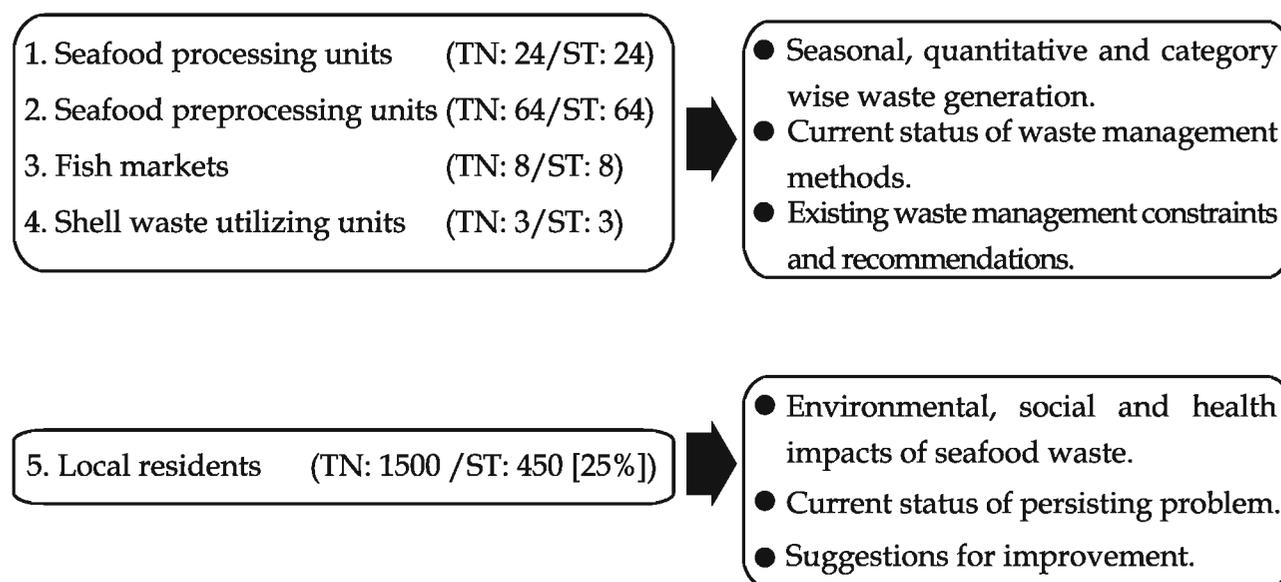


Figure 3.2. Survey unit categorization and primary data collection (TN-Total Number, ST-Sample Taken)

3.3.3.2 Secondary Data:

The secondary data were collected from published journals, government offices in the area, authority websites and published survey reports.

3.4 Results and Discussion

3.4.1 Current status of seafood raw material procurement and waste generation by the processing units

The combined Total Processing Capacity (TPC) of all the seafood processing units (Plate 3.3(a)) in ASIB is around 711 metric ton/day, currently handling only an average amount of 223 metric ton/day. The production during the post trawl ban period (from August to December) is 296.76 metric ton/day while during the pre-trawl ban period (from January to May), it is 191.95 metric ton/day and during trawl ban months (of June and July) it is about 116.182 metric ton/day. Among the raw material varieties processed in ASIB, shrimp accounted about 29167 metric ton/year, cephalopods 27907 metric ton/year and finfish 23296 metric ton/year. Among finfishes, tuna alone were processed at a quantity of around 16906.5 metric ton in the area. As far as the waste generation is concerned, shell waste contributes 10625 metric ton, cephalopod waste 5581.4 metric ton and finfish waste 6437 metric ton. Among the finfish waste generated in the area, tuna waste accounted around 4564.8 metric ton. Table 3.2 depicts the current status of seafood waste production from processing units in Aroor Seafood Industrial Belt. Different varieties of seafood wastes available in the study area are shown in Plate 3.3 (a-c).

Table 3.2 Current status of seafood waste production from processing units in Aroor Seafood Industrial Belt

Seafood waste production status/day			
Average amount of liquid waste generated/day (million L)	1.631		
Average amount of solid waste generated/day (t)	67.7		
Item wise production of solid waste/year (t)	Cephalopod	Shrimp	Fish
Pre trawl ban period (January-May)	1976.95	4124.59	2292.6
Trawl ban period (June-July)	471.5	1062.9	528.9
Post trawl ban period (August-December)	3132.93	5437.54	3615.6
Total amount of solid waste generated/year (t)	22643.4		
Total amount of liquid waste/year (million L)	534.7		

Source: Primary survey

The data on the Total Processing Capacity (TPC) of the seafood processing units in ASIB and their current average per day handling reveals that they are presently working only at 31 % of their total available capacity. The peak season of processing activity was found to be the post trawl ban period from August to December followed by the pre trawl ban period from January to May and minimum during trawl ban months of June and July. The cephalopods, shrimps and finfish formed the major seafood varieties currently being processed in the area. Shrimp dominates in quantity in ASIB followed by cephalopods and finfish. On the basis of a seasonal analysis, (Fig. 3.3) cephalopods exceeds shrimps in quantity during the post trawl ban period while shrimps dominate during the pre trawl ban and trawl ban seasons Tuna is emerging as a major seafood variety in the area, forming about 21% of total procurement annually.

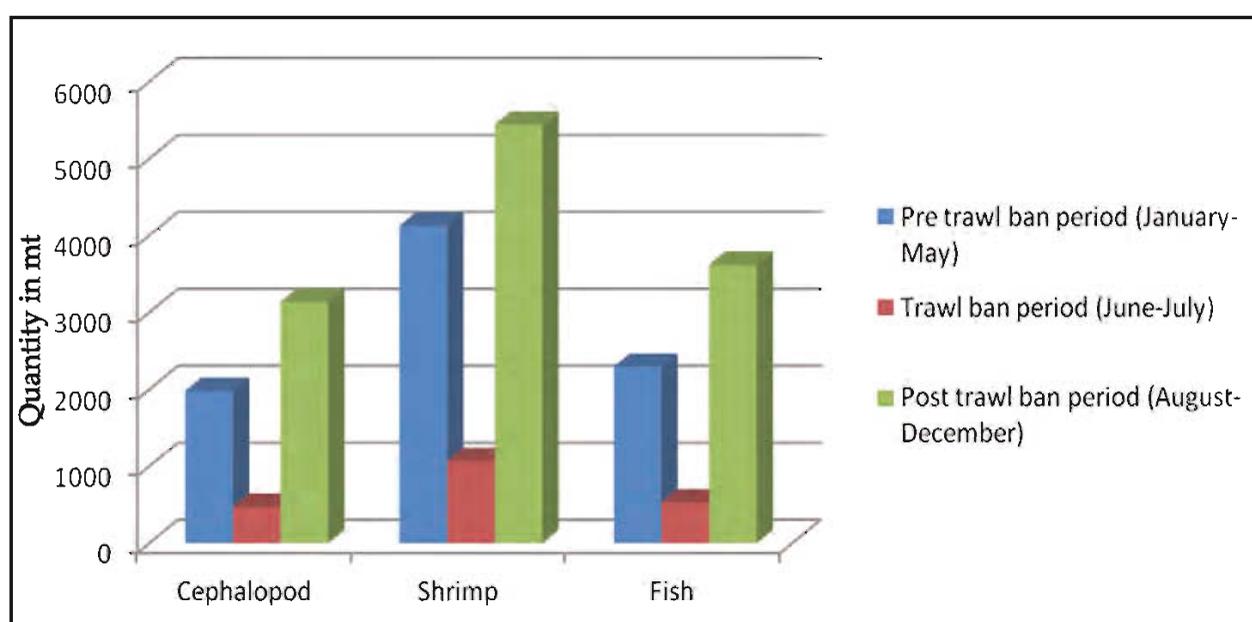


Figure 3.3: Seasonal production of seafood waste by the processing units in ASIB

In ASIB among the seafood waste generated by the seafood processing units, the maximum quantity was accounted by the shell waste followed by finfish waste and cephalopod waste. Tuna waste and finfish waste accounted for 20% and 71% respectively of the total seafood waste produced by the processing units in the area.

3.4.2 Current status of seafood raw material procurement and waste generation by the seafood pre-processing units

The pre-processing sector (Plate 3.3 (b) and (c)) in ASIB handles around 52829 metric ton of raw material per year. Shrimp is procured to the tune of 18735 metric ton/year, cephalopods 18109.6 metric ton/year and finfish around 15980 metric ton/year. About 208.678 metric ton of seafood was processed per day during post trawl ban period while during the pre-trawl ban season it was 137.515 metric ton/day. During trawl ban period, a quantity of 15 metric ton/day was reported to be processed. The annual production of shell waste, finfish waste and cephalopod waste were reported to the tune of 9181.6, 4380 and 4238 metric tons

Table 3.3 Current status of seafood waste production from pre-processing units in Aroor Seafood Industrial Belt

Seafood waste production status/day			
Average amount of liquid waste generated/day (million L)	0.89		
Average amount of solid waste generated/day (t)	69.2		
Item wise production of solid waste/year (t)	Cephalopod	Shrimp	Fish
Pre trawl ban period (January-May)	1638.56	3613.30	1711.75
Trawl ban period (June-July)	87.6	110.97	109.09
Post trawl ban period (August-December)	2511.63	5457.35	2559.4
Total amount of solid waste generated/year (t)	17799.6		
Total amount of liquid waste/year (million L)	22.4		

Source: Primary survey

The pre-processing sector in ASIB handled shrimp in highest quantity annually followed by cephalopods and finfish. The seasonal analysis showed that the maximum activity was during the post trawl ban season followed by the pre trawl ban season while the trawl ban period reported a comparatively lower quantum of processing. The first two seasons were dominated by shrimp and during the trawl ban season, cephalopods excelled followed by fin fish and shrimp. Shell waste dominated in quantity followed by finfish waste and cephalopod waste. Figure 3.4 depicts the seasonal production of seafood waste by the pre-processing units in ASIB.

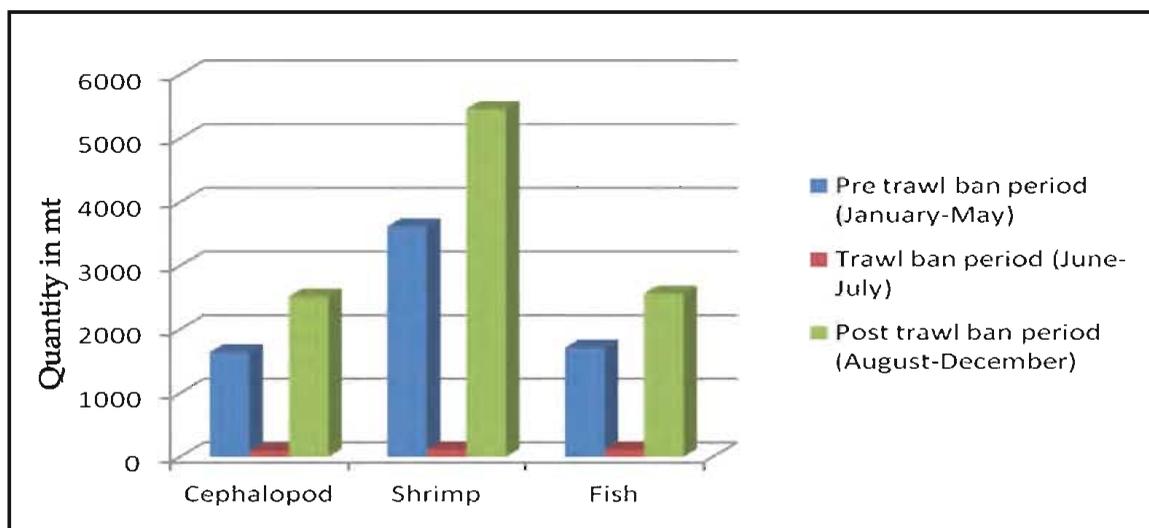


Figure 3.4: Seasonal production of seafood waste by the Pre-processing units in ASIB

3.4.3 Current status of seafood raw material procurement and waste generation by the fish markets and shell waste utilization by Chitin/Chitosan units

The fish markets in ASIB handle around 1353 metric tons of seafood annually, generating about 405.9 metric ton of seafood waste. The quantity handled during the post trawl ban months was around 676.5 metric ton/year while during the pre-trawl ban period and trawl ban period it was around 405.9 and 270.6 metric ton/year respectively. The seasonal waste production during the pre-trawl ban, trawl ban and post trawl ban periods were 121.8, 81.18 and 202.95 metric tons respectively. The Chitin/Chitosan units (CCU) procured about 7560 metric ton of shell waste annually. About 94.5, 37.8 and 94.5 metric ton of Chitin and 63, 25.2 and 63 metric tonnes of Chitosan were manufactured by the units during the pre-trawl ban, trawl ban and post trawl ban seasons respectively. This led to the generation of around 226.8 metric ton of solid waste and around 9 million L of liquid waste annually from the Chitin/Chitosan plants.

The maximum quantity of seafood handled by the fish markets in the area was during the post trawl ban months followed by the pre trawl ban and trawl ban periods. The same pattern was also observed in the waste production pattern during the pre-trawl ban, trawl ban and post trawl ban periods. The Chitin/Chitosan units (CCU) procured about 25.2% of shell waste generated in the area for by-product development.

3.4.4 Seafood effluent management

The most widely adopted effluent treatment technology adopted by the processing units (Table 3.4) in ASIB is the aerobic method involving chemical treatment and carbon/sand filtration. At an average around ₹ 15000 to ₹ 45000 is spent by each plant as operational cost for ETP per month. In the case of seafood pre-processing units, they adopted a 3 tank overflow method of sedimentation involving an initial investment cost of ₹ 10000 to ₹ 25000 with an average capacity of 1500 to 2500 L per day.

The results revealed that all the seafood processing units in ASIB are equipped with Effluent Treatment Plants (ETP) for discharging the effluent to the backwaters as followed by the qualitative specifications of the Kerala State Pollution Control Board (KSPCB) - General Standards for Discharge of Environment Pollutants: Effluent, Gazette Notification of MOEF, May 1993. This is in view of the fact that the waste water from seafood processing plants contains large amounts of organic matter, small particles of flesh, breaching, soluble proteins, chemicals and carbohydrates which can otherwise pollute the water body.

In the case of seafood pre-processing units, only 42.3% are having an ETP which is an alarming situation. This is because the number of pre-processing units varies in number from year to year depending on the availability of raw material in the area. Even though it is mandatory for the processing units to have a fully functioning pre-processing unit within the factory, most tend to outsource it in order to cope up with the labour expenses. The unregulated number of big and small pre-processing units without proper effluent treatment options tends to discharge non-treated waste water, high in nutrients to the nearby water resources, resulting

in health and social issues. Plate 3.4 depicts various stages in a processing/pre-processing unit generating liquid and solid wastes.

Table 3.4 Current status of effluent treatment technology adopted by processing units in Aroor Seafood Industrial belt

Effluent Treatment Plant Technology	Average Capacity (L/day)	Average Initial Investment Cost (INR)	Average Daily Operational Cost (INR)
Aerobic method with chemical treatment	10000 - 40000	57000 - 5,74,000	900 - 1000
Aerobic method with chemical treatment, carbon and sand filtration	15000 - 100000	1,70,000 - 11,50,000	1000 - 1700
Aeration and sedimentation	10000 - 50000	3,45,000- 17,23,674	600 - 1000

Source: Primary survey

The study revealed that about 22.7% of the ETPs installed by the processing units in ASIB are currently operating under capacity i.e. handling more liquid waste than the designed capacity, thus not leaving any other options but to discharge directly to the outlet. Even the units approved by national and international agencies tend to bend the rules by under operating the installed ETP. The high initial investment cost and daily operational costs of current ETP technologies are blamed by the processing units for this action. Figure 3.5 depicts the ETP technology preference by the processing units in ASIB.

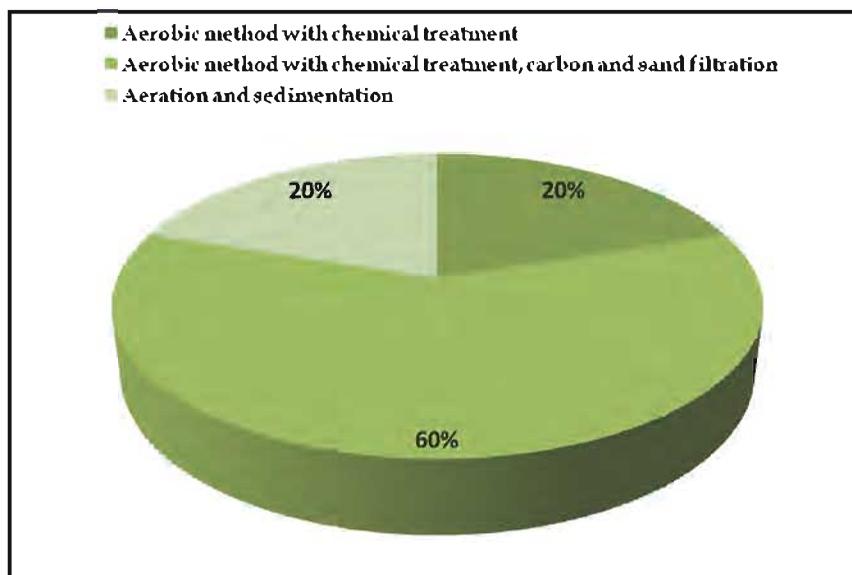


Figure 3.5: ETP technology preference by the processing units in ASIB

3.4.5 Seafood solid waste management

The entire quantity of solid waste generated in ASIB is disposed by the private contract system. Some part of the seafood waste generated also fetches a nominal income to the units. Finfish and cephalopod wastes are fetching a price of ₹35 to ₹40 per box (30-40 Kg) while the shrimp shell waste fetch around ₹50 to ₹60 per box. On the other hand, each unit has to spend around of ₹31 per box for disposal of unutilized seafood solid waste. The pre-processing units in ASIB also exhibited a similar pattern of seafood solid waste management. Current status of income generated from solid waste trading and expenditure incurred for solid waste movement by the fish processing and pre-processing units in Aroor Seafood Industrial Belt is given in Table 3.5.

Table 3.5 Current status of solid waste management by the fish processing and pre-processing units in Aroor Seafood Industrial Belt

	INCOME & EXPENDITURE STATUS	ARoor SEAFOOD INDUSTRIAL BELT	
		PU	PPU
	INCOME		
a.	Total quantity marketed for valorisation/year (t)	6862.11	6532.2
b.	Income/unit/day (Rupees)	1200	500
c.	Income/unit/year (Lakh Rupees)	3,98,800	1,48,700
d.	Total income by all units combined/year (Lakh Rupees)	95.7	95.2
	EXPENDITURE		
a.	Total quantity moved through private contract/year (t)	15781.3	20247.39
b.	Expenditure/unit/day (Rupees)	1700	900
c.	Expenditure/unit/year (Lakh Rupees)	5.85	2.81
d.	Total expenditure by all units combined/year (Lakh Rupees)	140	180

Source: Primary survey

PU-Processing units, PPU-Pre-Processing units

The shell waste utilizing units in ASIB entirely depended upon private contract for removal of their solid waste, which is thereafter dried for manure. In the case of fish markets

(Table 3.6), waste in the area is predominantly handled by the local authority followed by the local utilisation for livestock feeding. A significant quantity is noted to be dumped into local water bodies.

Table 3.6: Current status of seafood waste management methods adopted by Fish markets and Chitin/Chitosan units in Aroor Seafood Industrial Belt.

Seafood solid waste management method	FM	CCU
Back water/canal dumping (%)	15	0
Live stock feed (%)	20	0
Contract removal (%)	5	100
Authority removal (%)	60	0
Biogas plant (%)	0	0

Source: Primary survey FM-Fish markets, CCU-Chitin/Chitosan production units

The results reveal that the whole amount of solid waste generated in ASIB is disposed of by the private contract system. In ASIB, due to the increasing awareness on the nutritional quality of seafood and by-product valorisation possibilities as a result of the intervention by the Government and Non-Government R&D institutions in the area, a portion of the solid waste generated is sold to private parties for a nominal price. Around 20.41% of the cephalopod waste, 43.35% of finfish waste and 27.6% of shrimp shell waste are currently utilized for by product development. In the case of the seafood waste generated by the pre-processing units in ASIB, about 31.38% of cephalopod waste, 19.9% of finfish waste and 23.73% of shrimp shell waste is currently being marketed for valorisation. Figure 3.6 gives a comparison of income and expenditure incurred by the processing and pre-processing units in ASIB for solid waste management.

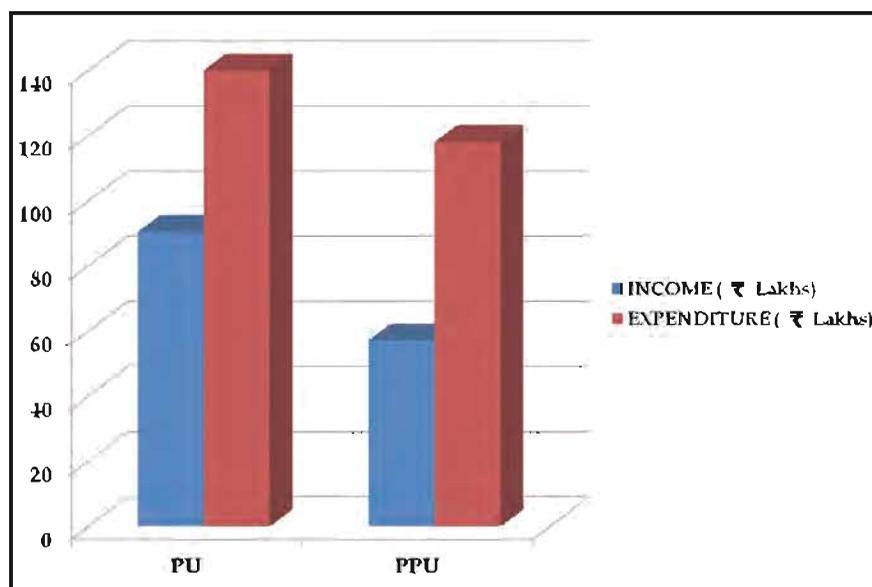


Figure 3.6: Comparison of income and expenditure (INR) incurred by the processing (PU) and pre-processing units (PPU) in ASIB for solid waste management.

In spite of the pressure exerted on the seafood processing units due to their proximity to highly populated and environmentally sensitive areas like backwater canals, a considerably large amount of waste is disposed as such which otherwise is cost involved. The dependency on private parties on this regard increases the risk as there is no proper authority control on the activity of these private parties. The solid waste in many cases is dumped in an irresponsible manner creating serious environmental and social problems. Figure 3.7 depicts the existing pattern of seafood waste management in Aroor seafood industrial belt.

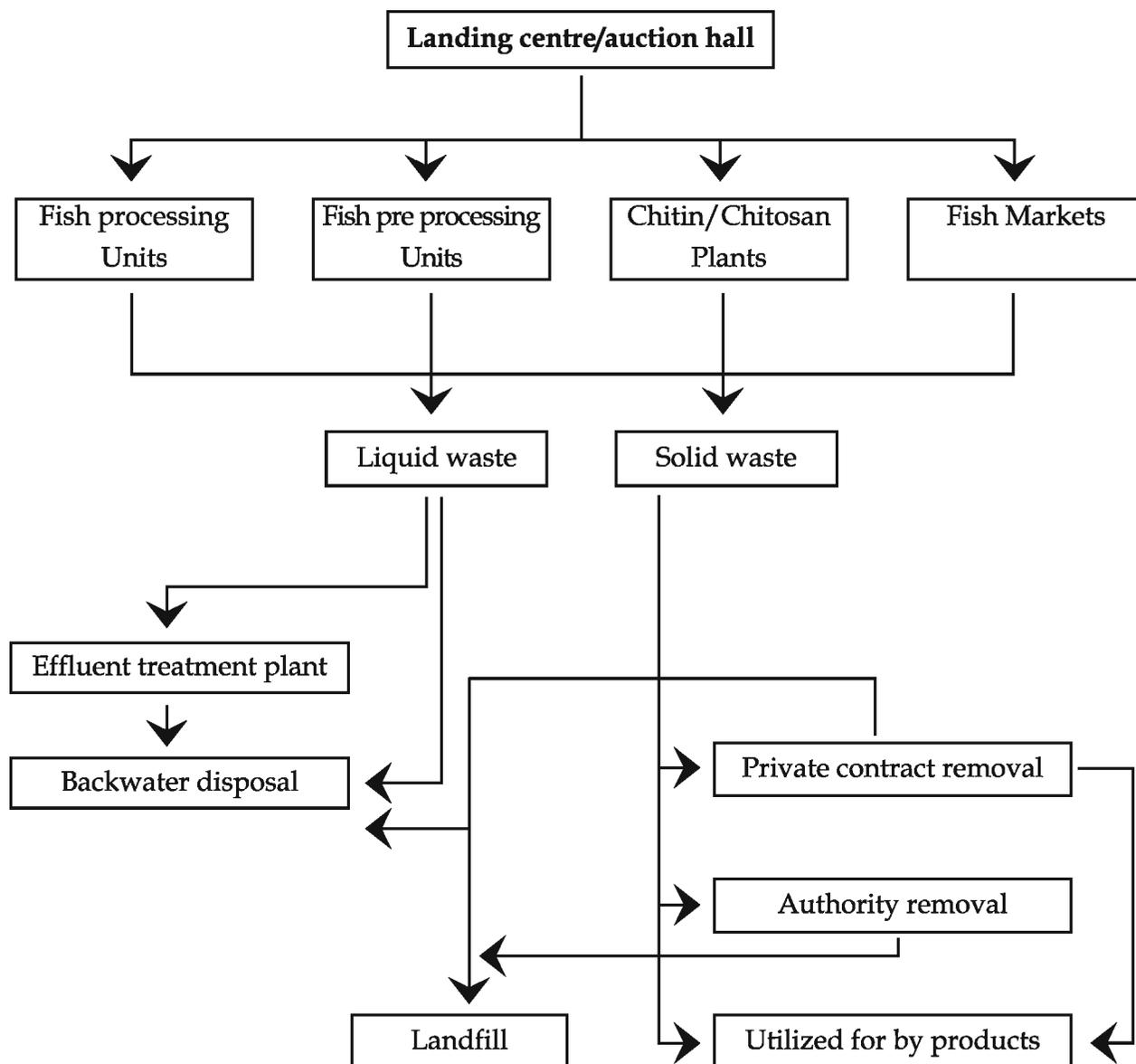


Figure 3.7. Existing pattern of seafood waste management in Aroor Seafood Industrial Belt

3.4.6 Issues related to seafood waste management in ASIB

The awareness percentage on seafood waste utilization methods among the processing units in ASIB was found to be 62.5%. About 38% preferred an outside agency for

handling the seafood by-product development activities if suitable technologies are transferred, 62% agreed to do it in plant if profitable. About 20% of the units were concerned of the cost involved for adopting seafood waste utilization technologies while 27% expressed lack of sufficient space for adopting any method for seafood waste utilization. The seafood waste utilization method awareness among the pre-processing units was found to be 100%. About 73.1% preferred an outside agency for handling the seafood by-product development activities and around 26.9% agreed to do it in plant if profitable. Lack of space and cost involved were the most expressed concerns for adopting methods of seafood waste utilization. In most of the fish markets in the study area, lack of space for handling seafood waste is the problem of highest priority to be tackled. And most of the beneficiaries recommended a common waste treatment facility to be installed in the market space. About 85% are currently aware of the seafood waste utilization methods available.

All of the Chitin/Chitosan units expressed smell as the largest problem encountered in waste management and recommended a common treatment facility for waste treatment. All of them indicated lack of technology for utilizing the solid and liquid waste generated and would prefer to install it in plant if any technology is transferred.

The awareness percentage on seafood waste utilization methods among the processing units in ASIB revealed in the study was not as promising as expected. The study emphasised on the need for more intervention from the authorities on the matter in the form of awareness campaigns and technology popularisation efforts, which is more important in the light of the fact that a higher percentage is ready to implement the technology within the premises of the unit if successfully transferred. It was also evident that a majority of units were concerned about the availability of space for adopting such technologies rather than the costs involved.

The seafood waste utilization method awareness among the pre-processing units was found to be more promising. But in their case, the preference for an outside agency for taking up such activities could be well understood considering the limited amount of space and investment available. Like the processing units, lack of space and cost were the most expressed concerns for adopting the same. For the fish markets on the other hand, space was the biggest concern and wanted a common treatment facility for the same while being highly aware about the valorisation options. The largest concern for the Chitin/Chitosan units in the area was the smell as they have to deal with shell waste. They also need to be made aware of the technologies available for utilising the solid and liquid waste generated after the treatment processes for further valorisation.

In the case of the responses collected regarding the seafood waste management difficulties faced by the processing (Table 3.7) and pre-processing units (Table 3.8) in the area, environmental pollution was mentioned as the biggest challenge to be faced by the units while going for waste management. Delay in timely waste collection by the authorities and the private parties envisaged for the purpose, was the second largest hurdle. Even though so many points

were raised, the striking factor was that a large percentage of units refused to give any comments on the matter, pointing out into the serious social and legal scenario prevailing in the study area with regard to seafood waste management.

Table 3.7 Response on seafood waste management difficulties and recommendations by processing units in Aroor Seafood Industrial Belt

Response on seafood waste management difficulties	Percentage
1 Environmental pollution	17.4
2 Cost intensity	8.7
3 Lack of an organized waste management system	4.35
4 Delay in timely waste disposal	13
5 Lack of sufficient contract removal agents	4.35
6 Frequent ETP failure	4.35
7 Increased difficulty in rainy season	8.7
8 No comments	39.15
Response on seafood waste management recommendations	Percentage
1 Common treatment facility	25
2 Development of new by-products from seafood waste	62
3 No response	13

Source: Primary survey

Table 3.8 Response on seafood waste management difficulties and recommendations by Pre-processing units in Aroor Seafood Industrial Belt

Response on seafood waste management difficulties	Percentage
1 Cost intensity	3.85
2 ETP malfunction	3.85
3 Delay in waste collection	7.7
4 Lack of suitable effective technology	3.85
5 Water pollution	11.5
6 No Comments	69.25
Response on seafood waste management recommendations	Percentage
1 Common treatment facility	16.7
2 Development of byproducts	63.34
3 No comments	3.33
4 Training in fish waste management and utilization	3.33
5 Improved effluent treatment technology	3.3
6 Credit support from Government agencies	10

Source: Primary survey

3.5 Impact of seafood waste on coastal livelihood: A case study

Chandiroor canal (Fig. 3.8) located in the Chandiroor Panchayath in ASIB is a typical instance of impact of seafood waste pollution on the normal living conditions of the local population and the environment. The canal extends to a length of 1.4 Km and a width of 12 meters. It is on both sides connected to back waters. A number of seafood processing and pre-processing units have their outlets directly opening to the canal. In Chandiroor Panchayath around 300 households are directly affected by the ill effects of canal pollution.

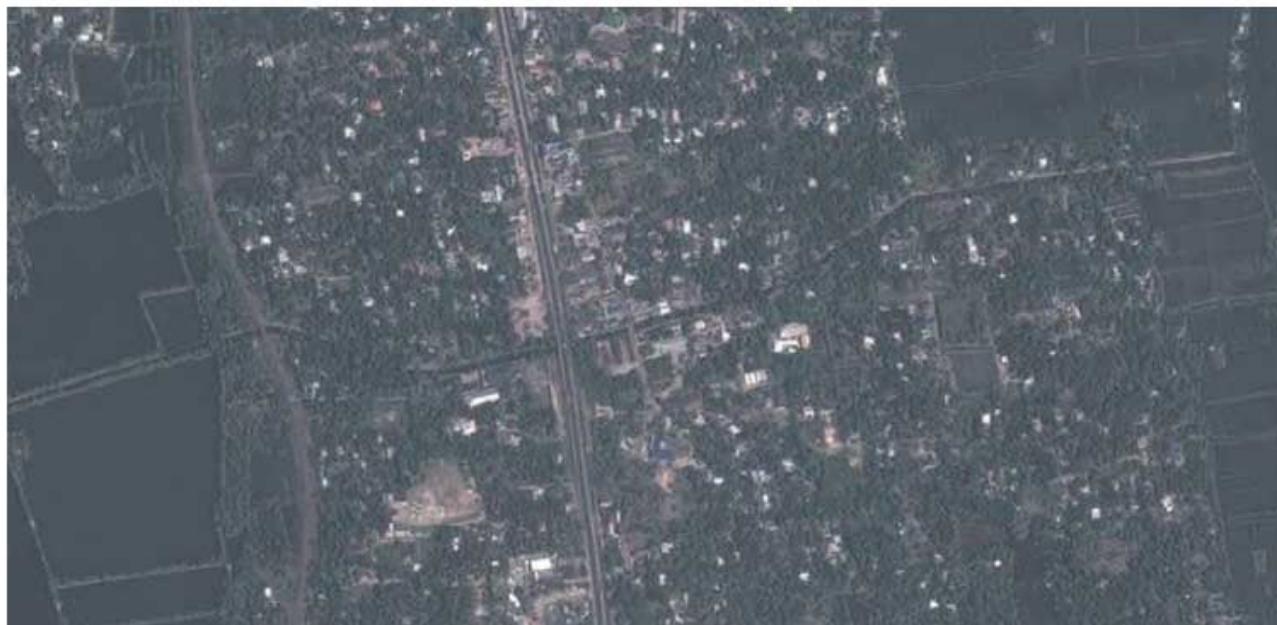
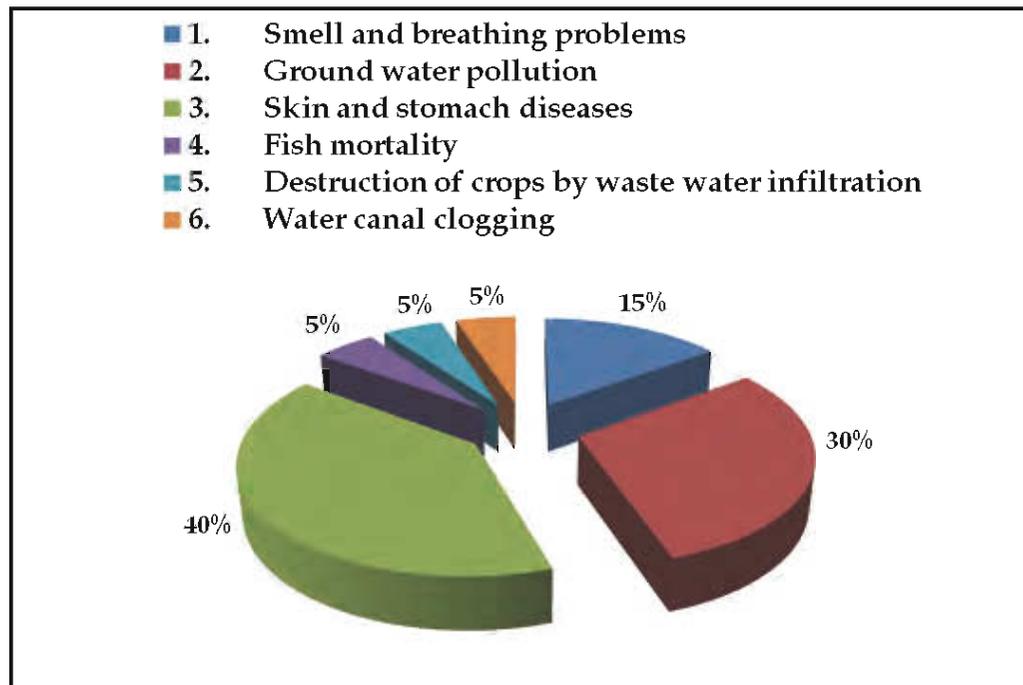


Figure 3.8: Satellite image showing the location of Chandiroor canal. (Source: Wikimapia.com)

The issue of pollution from seafood effluent discharge to the Chandiroor Canal is persisting for about 15 to 25 years and the region has been exposed to a series of violent agitations with little effect. The Chandiroor village was declared as the first Siddha Village in India as a part of the village adoption program of Santhigiri Siddha Medical College, Thiruvananthapuram, Kerala in 2006. A survey conducted (Santhigiri Siddha Medical College, Medical Survey, 2006.) by the medical college revealed that the inhabitants were suffering from rheumatic complaints, respiratory ailments, skin disorders, diabetes mellitus, piles, hypertension etc. Probable reasons given for the above said diseases were water contamination from ice plants, prawn peeling and coconut husk processing units; water stagnation which breeds mosquitoes; lack of sanitation facilities and, personal hygiene and lack of pure drinking water facilities. The percentage response by the local residents on seafood waste management recommendations and pollution issues of Chandiroor canal in Aroor Seafood Industrial Belt is depicted in Fig. 3.9.

Even though the residents raised the necessity for a common waste treatment facility as a prime focus, the suggestion for allocating a nonresidential area for such facility, raised by a much lesser percent of people, is actually coming in each other's way in the area. The difficulty in finding such an area in a highly populated coastal village has pushed the authorities'

attempts without any fruitful results. The health issues revealed in the study is in sync with the studies conducted by the Santhigiri Medical team in the area, justifying the ill effects of seafood waste pollution. The seafood waste management will be successful only if the ransom quantity of waste could be redirected to a channel through recycling. The succeeding chapters deal with practical solutions for the problem of seafood waste management.



Source: Primary survey

Figure 3.9: Seafood waste pollution issues raised by the local residents of Chandiroor.

3.6 Conclusion

- The seafood processing units in ASIB are presently working only at 31 % of their total available capacity.
- The peak season of processing activity is the post trawl ban period from August to December followed by the pre trawl ban period from January to May and minimum during trawl ban months of June and July.
- The cephalopods, shrimps and finfish formed the major seafood varieties processed in the area.
- Tuna is emerging as a major seafood variety in the area forming about 21% of total procurement annually.
- Among the seafood waste generated by the seafood processing units in ASIB, the maximum quantity was accounted by the shell waste followed by finfish waste and cephalopod waste.
- In the case of seafood pre-processing units, only 42.3% were having Effluent Treatment Plants (ETP).

- In the case of seafood pre-processing units, only 42.3% were having Effluent Treatment Plants (ETP).
- The case study reveals that the seafood pollution not only exerts environmental impacts but also livelihood and health issues among the coastal community.



Show-cause notice served on Chief Secretary

Saturday, Nov 22, 2003

THE  HINDU

KOCHI Nov. 21. A Division Bench of the Kerala High Court issued notice to the Chief Secretary and the Alappuzha District Collector asking them to show cause why contempt of court proceedings should not be initiated against them for their failure to comply with an earlier directive. The High Court had directed the respondents including the Chief Secretary and the District Collector to take steps for setting up a common effluent treatment plant at Chandiroor in Cherthala to check the pollution caused by peeling sheds.

<http://www.hindu.com/2003/11/22/stories/2003112208240400.htm>

Call to minimise pollution

Friday, Dec 17, 2004

THE  HINDU

ALAPPUZHA, DEC. 16. Participants of a media seminar on the 'Total Sanitation Campaign' in Alappuzha district organised by the Alappuzha district sanitation committee here today have called for enlarging the scope of the total sanitation campaign to minimise various types of pollutions. Some of the participants noted that residents of Aroor panchayat in the district, where a large number of seafood processing factories exist, were facing a threat to their existence because of the large amount of wastes being produced by the factories.

<http://www.hindu.com/2004/12/17/stories/2004121708110300.htm>

Peeling sheds polluting water: PCB

Wednesday, Mar 30, 2005

THE  HINDU

ALAPPUZHA, MARCH 29. An inspection conducted by the officials of the State Pollution Control Board (PCB) at the prawn peeling sheds and meat processing centres in and around Kakkazham, near Ambalappuzha in the district found that they were polluting the Pappithode, a canal there, and other water bodies in the area.

<http://www.hindu.com/2005/03/30/stories/2005033003560300.htm>

Diseases Follow Environmental Degradation

By Archana Devraj *Tierramérica Issue of December, 02, 2006*

The contamination of canals and other sources of water led to the outbreak of chikungunya in the Indian state of Kerala. The finger of blame is pointing at fish processors and coconut fiber factories. CHERTHALA, India, Dec 2 (Tierramérica)- To find the origin of the chikungunya epidemic, which has already claimed 125 lives in the Indian state of Kerala, take a look at some of the area's water sources, which have been turned into vast pools of industrial runoff. "It is not surprising that chikungunya has hit Cherthala. The pollution level in the canals is very high. In fact, a report of the State Pollution Control Board in 2002-03 had described Aroor in Cherthala as the most polluted 'panchayat' (village) in the state. Similarly, the Chandiroor canal flowing through the area had been described as the most contaminated," says journalist C Radhakrishnan, who lives in Cherthala, and was among the first to report the chikungunya outbreak. According to the local residents, nearly 100 big and small marine products exporting units dot the northern parts of Cherthala. The waste from the processing units and the peeling sheds is dumped into the canals leading to a choking of the water flow.

<http://www.tierramerica.info/print.php?lang=eng&idnews=253&olt=43>

Closure notice to 24 seafood processing units

Thursday, January 04 2007



Thiruvananthapuram, Jan 4 (UNI) The Kerala State Pollution Control Board has issued closure notice to 24 seafood export-related industrial units in the Aroor-Chandiroor Area of Alappuzha District for 'not following' the Board's guidelines on pollution control. "Following a High Court order based on a PIL that prawn processing units in the area are releasing large quantity of effluents into badwaters, we have inspected about 50 units and found 24 of them are not following the guidelines," Board Chairman G Raja Mohan told UNI.

<http://news.oneindia.in/2007/01/04/closure-notice-issued-to-24-seafood-processing-units-1167898217.html>

Plate 3.1: Media reports on environmental crisis around ASIB



3.2(a): A modern processing Unit



3.2(b): A traditional pre-processing unit



3.2(c): A modern pre-processing unit



3.2(d): A busy fish market in the study area.

Plate 3.2: Potential seafood waste generation units in ASIB



3.3 (a): Cephalopod processing waste



3.3 (b): Shrimp shell waste

(Source: <http://shrimpmeal.files.wordpress.com/2012/07/dried-shrimp-heads.jpg>)



3.3 (c): Fin fish waste

(Source: <http://mudpreacher.files.wordpress.com/2012/11/fish-guts.jpg>)

Plate 3.3: Different varieties of seafood waste available in the study area



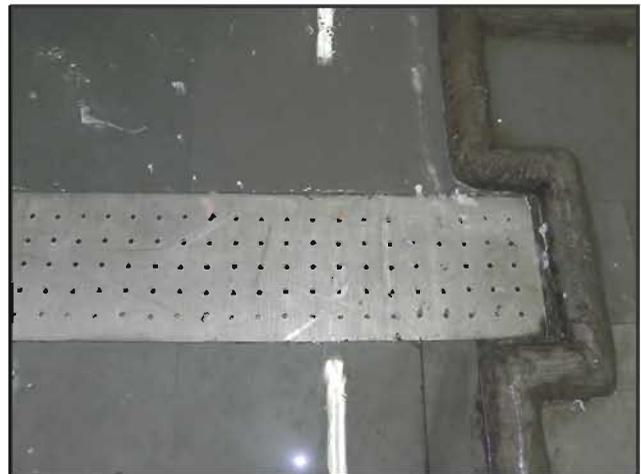
3.4(a): Raw-material being weighed for processing



3.4(b): Liquid and solid wastes being separated



3.4(c): Solid waste ready for disposal



3.4(d): Drain inside the processing hall



3.4(e): Drain outside the processing hall



3.4(f): An effluent treatment plant

Plate 3.4: Various stages of liquid and solid waste generation in seafood processing

CHAPTER 4

BIOCHEMICAL ANALYSIS OF SEAFOOD PROCESSING WASTE

Contents	4.1 Introduction
	4.2 Materials and methods
	4.3 Results & Discussion
	4.4 Conclusion

4.1 Introduction

The seafood processing, pre-processing, waste utilisation and marketing industries in ASIB produce around 711 metric tonnes of solid waste on a daily basis. In order to formulate a plan for effective utilisation of the waste, a basic knowledge about the quality is required. The essential data on biochemical quality of different varieties of seafood waste generated in the zone could be utilised for designing a Waste Utilisation Index (WUI)(Appendix 2). This chapter attempts to classify different varieties of seafood waste generated in the area and to analyse the basic biochemical quality parameters such as proximate composition, amino acid profile, fatty acid profile and chitin content, so as to create a WUI, based on the data available.

Analysis of biochemical composition of fish flesh is considered to be a good indicator for the fish quality (Hernandez *et al.*, 2001). Biochemical parameters like moisture, dry matter, protein, lipids and minerals are considered to be the most significant components of nutritive value in seafood (Steffens, 2006). Determination of proximate composition is the primary step towards biochemical profiling of any nutritionally significant food. Waterman (2000) stated that the quantification of proximate composition is vital in ensuring the necessities of food regulations and commercial standards. Sutharshiny and Sivashanthini (2011) also considered proximate composition analysis as the most important aspect in fish nutrition studies. Moisture content is considered to be a good indicator by many researchers with its relative content of energy, protein and lipid (Aberoumad, 2012). Among the proximate parameters, protein is of primary importance, because of the amino acid composition and degree of digestibility (Louka *et al.*, 2004). The importance of protein analysis in food industries like seafood has been emphasized by researchers like Rustard (2010a). The value and the quality of the protein in the raw material are determined by the content and the properties of the protein the raw material holds (Owusu-Apenten, 2002).

Another important nutritional factor in seafood is the lipid content. Marine lipids are rich in polyunsaturated n-3 fatty acids (PUFA) such as docosahexaenoic acid (DHA; 22:6 n-3)

and eicosapentaenoic acid (EPA; 20:5 n-3) (Rustard 2010b). The PUFA are significant for having medicinal properties like preventing coronary heart diseases and positively effecting on brain activity as well as stimulating immune system (Boissonneault, 2000 and Narayan *et al.*, 2006). Chitin, present in the shell waste of the crustaceans is another important biochemical parameter to be assessed for commercial utilization. Chitin is the major structural component of the exoskeleton of shellfish (Ornum, 1992). Chitin and its derivatives are commercially important with the properties such as non-toxicity, non-allergenicity, anti-microbial effect, insolubility in water and resistance to acids, alkalis, and many organic solvents and having flexible biological activity and exceptional biocompatibility.

4.2 Materials and methods

4.2.1 Sample collection and preparation

The seafood processing waste samples were collected from various Waste Generation Points (WGP) in the study area, such as fish markets, pre-processing units and processing units. The samples were stored in ice and brought to laboratory. The samples were homogenised to uniform texture, placed in polythene bags and stored at -20°C until used for extraction and analysis. For all analysis, samples were taken in triplicate. The seafood wastes were broadly classified as given below and an analytical plan was prepared for assessing the quality (Table 4.1).

4.2.2 Proximate composition analysis

4.2.2.1 Determination of moisture

The moisture content was estimated by the method of AOAC (2000). The moisture content was determined by drying 10 g sample at 103°C in a thermostatically controlled hot air oven. The samples were taken in a pre-weighed petri dish and kept in oven and the reduction in weight was checked by repeatedly heating and then cooling the sample in desiccator till the weight became constant. Moisture content was determined as percentage.

4.2.2.2 Determination of Protein

1 g of homogenized sample was accurately weighed into a digestion tube. About 2 g of digestion mixture and 10 ml of concentrated sulphuric acid were added to the sample taken in the digestion tube. The samples were digested to a clear solution in a KEL 12 PLUS EK digestion unit and made up to 100 ml with distilled water. 5 ml of the digest was pipetted out into the Kjeldahl Micro distillation apparatus. 10 ml of 40% NaOH was added to the sample in the distillation unit to make it alkaline. The bottom end of the condenser was fitted to a delivery tube immersed in 10 ml of 2% boric acid with added Tachiro's indicator. The ammonia produced on steam distillation was absorbed into the boric acid solution. The distillate collected was back titrated against N/70 sulphuric acid. The total nitrogen content was calculated and the crude protein content (%) in the sample was calculated by multiplying the percentage of nitrogen content by the factor 6.25.

Table 4.1: Analytical plan for assessing the quality of seafood wastes

	Proximate composition				Amino acid profile	Fatty acid profile	Chitin content
	Moist ure	Crude protein	Crude fat	Ash content			
Tuna (<i>Thunnus albacares</i>)							
Tuna Head waste	√	√	√	√	√	√	×
Tuna Viscera	√	√	√	√	√	√	×
Tuna Dark Muscle (Raw)	√	√	√	√	√	√	×
Tuna Dark Muscle (Pre-cooked)	√	√	√	√	√	√	×
Fish head & viscera							
<i>Sardinella longiceps</i>	√	√	√	√	√	√	×
<i>Rastrelliger kanagurta</i>	√	√	√	√	√	√	×
<i>Nemipterus japonicus</i>	√	√	√	√	√	√	×
Cuttlefish processing waste							
<i>Sepia pharonis</i>	√	√	√	√	√	√	×
Squid processing waste							
<i>Loligo duvaccelli</i>	√	√	√	√	√	√	×
Shrimp head waste							
<i>Metapenaeus dobsoni</i>	√	√	√	√	√	√	×
<i>Parapenaeopsis stylifera</i>	√	√	√	√	√	√	×
<i>Penaeus indicus</i>	√	√	√	√	√	√	×
<i>Aristus alcockii</i>	√	√	√	√	√	√	×
Shell waste							
<i>Metapenaeus dobsoni</i>	×	×	×	×	×	×	√
<i>Parapenaeopsis stylifera</i>	×	×	×	×	×	×	√
<i>Penaeus indicus</i>	×	×	×	×	×	×	√
<i>Aristus alcockii</i>	×	×	×	×	×	×	√
Reference	AOAC (2000)	AOAC (2000)	AOA C (2000)	AOAC (2000)	Shang and Wang (1996)	AOAC (2000)	Spinelli et al. (1974).

4.2.2.3 Determination of Crude Lipid

Fat content of the moisture free sample was determined by extracting the fat by using a suitable solvent (Petroleum ether, boiling point: 40-60°C) by soxhlet extraction method (AOAC, 2000). About 2g of the moisture free sample was accurately weighed into an extraction thimble and was placed in the extractor. The extractor was connected to a pre weighed dry receiving flask and water condenser. The unit was placed on a water bath and temperature was maintained at 40°C - 60°C so that solvent boiled continuously and siphoned at a rate of 5-6 times/hr. The extraction was continued till the solvent in the extractor became colourless and fat free. The solvent in the receiving flask was evaporated completely and weighed for fat content.

4.2.2.4 Determination of Ash

The ash content was determined by incineration of the sample according to AOAC (2000). 2 g of moisture free sample was taken in a pre-weighed clean dry silica crucible and charred on low heat. It was then kept in a muffle furnace at 550°C to get a white ash that was cooled in desiccator and weighed.

4.2.3 Amino acid composition analysis by HPLC: (PICO -TAG method)

Quantitative determination of the amino acids was conducted using a Reverse Phase -High Performance Liquid chromatography (RP - HPLC) after pre-column derivatization using PITC (phenyl isothiocyanate) according to the method of Shang and Wang (1996).

4.2.3.1 Reagents

All reagents used were of HPLC grade.

(a) PICO - TAG Eluent A

Pico-Tag Eluent A was prepared by dissolving 19.0 g sodium acetate trihydrate in one litre of deionized Milli Q water. Added 0.5 ml triethylamine (TEA) to the above solution and titrated to pH 6.4 using glacial acetic acid. To the resulting solution added 63.8 ml of acetonitrile and filtered using sterile membrane filters (cellulose nitrate, 47mm Dia, 0.45 µm pore size, WCN type, Whatman Ltd, Maidstone, England).

(b) PICO - TAG Eluent B

Pico-Tag Eluent B was prepared by mixing HPLC grade acetonitrile and deionized milli Q water in the ratio 6: 4 by volume.

(c) Sample diluent solution

The diluent solution was prepared as follows: 710 mg of Na₂HPO₄ was added to one litre of water and titrated to pH 7.4 with 10 % phosphoric acid (H₃PO₄). The resulting solution was mixed with HPLC grade acetonitrile, so that acetonitrile was 5 % by volume.

(d) Sample redrying Solution

The redrying solution consisted of HPLC grade methanol, millipore water, and triethylamine (TEA) in the ratio 1:1:0.5 by volume.

(e) Sample derivatization Solution

The derivatization reagent consisted of HPLC grade methanol, triethylamine, millipore water, and phenyl-isothiocyanate in the ratio 7:1:1:1 by volume.

4.2.3.2 Sample preparation for Pico-Tag amino acid analysis

Samples (100 – 200 mg tissue) were hydrolysed using 10.0 ml of 6 M HCl containing 2 % phenol at 110°C for 24 hours in heat sealed tubes, after flushing with nitrogen. The hydrolyzed sample solutions thus obtained were subjected to derivatization using PITC. Briefly, 5µl each of the above filtered sample solutions were transferred to a 1.0 ml sample tube and dried under vacuum to remove all traces of HCl. The samples in the tubes were treated with 20 µl of re-drying solution (Reagent 4.2.3.1 (d)), by gentle vortexing and dried under vacuum. Subsequently, the amino acids were derivatized by addition of 10µl of derivatization solution (Reagent 4.2.3.1(e)), to the above dried samples by gentle vortexing for 20 minutes at room temperature. The sample derivatization solution was further removed by drying under vacuum for 30 minutes. After derivatisation, one ml of sample diluent solution (4.2.3.1(c)) was added and mixed by vortexing for a few seconds and filtered using 0.45µm nylon filters (Whatman, Maidstone, England). For chromatographic analysis 10µl each of the above samples were injected into the HPLC.

4.2.3.3 Apparatus

HPLC system (Waters Model 2487) equipped with a Binary pump model M 515, a 600 Gradient mixer solvent delivery system and a 5 µm Pico-Tag Reversed Phase column (3.9 mm i.d. x 150 mm length) was used for the analysis of amino acids. The equipment is provided with column oven (TCM Waters), a dual λ absorbance detector (UV/VIS Model 484) and a manual injector. Data analysis was performed using EMPOWER 2 chromatography software.

4.2.3.4 Chromatographic conditions

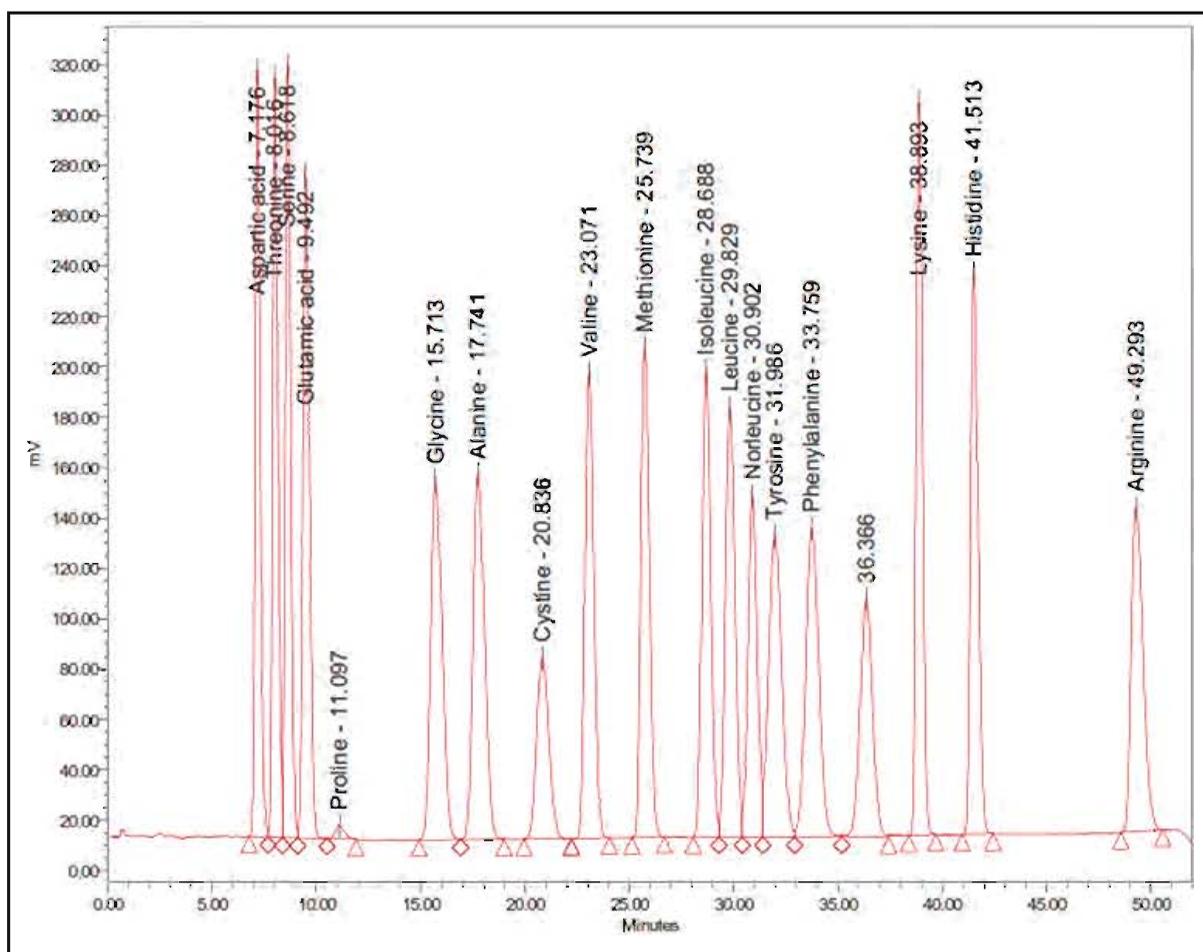
Chromatographic separation made use of continuous gradient elution with Pico Tag eluent A and Pico Tag eluent B for hydrolysate amino acid analysis. The gradient started at 100% eluent A and was decreased to 54% and finally increased to 100% in 14.5 min. The total separation time was less than 14 min and the gradient was run for 22.5 min to ensure full separation. Detection was monitored at 254 nm. The concentrations of all the standard amino acids were 2.5 µmoles/ml, except for cysteine with 1.25 µmoles/ml. The peak areas of individual amino acids were compared with standards and concentration was calculated after injecting 20 µl amino acid solutions. HPLC gradient profile developed for the separation of amino acids is given in Table 4.2 . Fig. 4.1 depicts the auto- scaled chromatogram of amino acid standards.

4.2.4 Extraction of total lipids

The total lipid of the tissues was extracted by the method of Folch *et al.* (1957). A weighed amount of the tissue was subjected to lipid extraction using chloroform-methanol mixture (2: 1).

Table 4.2: HPLC gradient profile developed for the analysis of amino acids.

Sl. No.	Flow (mL min ⁻¹)	Time (min)	% Eluent A	% Eluent B	%C	Curve
1	1.00	0.01	100.0	0.0	0.0	6
2	1.00	2.00	100.0	0.0	0.0	5
3	1.00	12.00	54.0	46.0	0.0	5
4	1.00	12.50	0.0	100.0	0.0	6
5	1.00	13.50	0.0	100.0	0.0	6
6	1.50	14.00	0.0	100.0	0.0	6
7	1.50	14.50	100.0	0.0	0.0	6
8	1.50	22.00	100.0	0.0	0.0	6
9	1.00	22.50	100.0	0.0	0.0	6

**Fig. 4.1 Amino acid standards: Auto- Scaled Chromatogram**

The extraction was repeated twice with fresh aliquot of chloroform-methanol mixture. The lipid extracts were transferred to a separating funnel and added 20% of water into it and left overnight. Next day the lipid extracts were drained through filter paper containing anhydrous sodium sulphate and was collected in round bottom flask and was evaporated to dryness in a flash evaporator. The lipid in the round bottom flask was made up to 10 ml using chloroform. From this 1 ml was taken into a pre-weighed vial and allowed to dry in warm temperature to constant weight and total lipid content was calculated from the difference in weight. From the above chloroform extract, 1 ml each was used for the estimation of various lipid components viz., cholesterol (total and free), triglycerides, free fatty acids and phospholipids after evaporating the solvent in air at room temperature.

4.2.5 Analysis of fatty acids

Fatty acids were analyzed according to the method of AOAC (2000). Methyl esters of fatty acids (FAME) were prepared and detected by gas chromatography.

4.2.5.1 Reagents

- 1 Boron Trifluoride reagent (BF₃).
- 2 Methanolic Sodium Hydroxide solution: 0.5N.
- 3 Petroleum ether.
- 4 Sodium Sulphate.

4.2.5.2 Procedure

A known weight of lipid, extracted by Folch method was taken into a round bottom flask and evaporated off the chloroform. 6 ml of methanolic NaOH and boiling chip were added. Attached condenser, and refluxed under nitrogen until fat globules disappear (usually 5-10 min). Added 6-7 ml BF₃ solution and continued boiling for 5 min. Removed from heat, and added 6 ml saturated NaCl solution. Stopped the flask and mixed vigorously for 15 sec while solution was still warm. It was then transferred into a 250 ml separating funnel. Washed the RB flask with 30 ml distilled water and transferred it into a separating funnel. Added 25 ml petroleum ether (PE) (b.p 60-80°C) to this separating funnel, shaken thoroughly and left for 5 min under nitrogen for separation. The lower aqueous layer was transferred to a round bottom flask and upper PE layer to another separating funnel. Lower aqueous layer in the round bottom flask was extracted twice with 25 ml of PE, and the upper PE layer was pooled with the above one in the separating funnel. The combined PE extracts were washed thrice with 20 ml portions of H₂O, collected the upper PE layer, filtered it through anhydrous sodium sulphate, and evaporated off solvent under vacuum. Made up the contents into 1 ml with PE and transferred into small vials flushed with N₂ and kept in a freezer till analysis.

Methyl esters of the fatty acid thus obtained were separated by gas liquid chromatography (Thermo Trace GC Ultra) equipped with a capillary column (30 m long and 0.54 mm diameter) and a flame ionization detector in the presence of hydrogen and air. The carrier gas was nitrogen

and the flow rate was 0.8ml/min. The initial temp was set as 70°C and was increased 3°C/min until a temperature of 250°C was obtained. Injector and detector temperatures were kept at 260°C and 275°C respectively. Fatty acids separated were identified by comparison of retention times with standard fatty acid methyl esters. Measurement of peak areas and data processing were carried out by Thermo Chrom card software. Individual fatty acids were expressed as a percentage of total fatty acids.

4.2.6 Analysis of Chitin content

The Chitin content was determined by the method of Spinelli *et al.* (1974). One gram of moisture free sample was digested with 100 ml of 2% NaOH at 100°C for 1 hour. The digested material was filtered through a coarse sintered glass and residue was digested again with alkali and filtered. The residue was treated with 100 ml of 5% HCl at room temperature for 15 hours, filtered and washed with hot distilled water. The washed residue was dried at 100°C for 6 hours and weighed and chitin content determined. Nitrogen content in the Chitin obtained was determined by using the Kjeldahl method (AOAC, 2000). True protein content was calculated by the formula,

$$\text{True Protein} = (\text{Total Nitrogen} - \text{Chitin Nitrogen}) \times 6.25$$

4.3 Results & Discussion

4.3.1 Proximate composition

Among the different groups of seafood waste varieties analysed for proximate composition (Table 4.3), the tuna waste showed maximum values for protein and fat. The tuna dark muscle (pre-cooked) and tuna dark muscle (raw) which were the contributions of the canning industry in the area showed high values in protein percentage. Studies conducted by Murthy *et al.* (2012) and Nguyen *et al.* (2011) indicated a similar pattern for tuna processing waste. The high protein level in pre-cooked tuna dark muscle was due to the low levels of fat, otherwise high in raw condition. Tuna head and viscera were high in crude fat content. In tune with findings of Zahar *et al.* (2002) and Rai *et al.* (2012) along with Nisha and Asadullah (2011), the fin fish head and viscera waste available in the area, of oil sardine, mackerel and Japanese sea bream, also showed a high percentage of protein while oil sardine waste reported an elevated level of crude fat content. Table 4.3 depicts the percentage composition of proximate components in these seafood wastes.

On the other hand, contradictory to the findings of Uddin *et al.* (2010) and Kechaou *et al.* (2009), the cuttlefish and squid processing waste showed a high moisture content and comparatively low protein and fat content. In the case of shrimp head waste available, *Penaeus indicus* showed a high level of crude protein content while the *Aristusalcokii* head waste reported the highest percentage of fat and *Metapenaeusdobsoniia* high level of ash content. These results were in tune with the findings of Ravichandran *et al.* (2009) and Sindhu and Sherief (2011).

Table 4.3: Proximate composition of seafood wastes

	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
Tuna (<i>Thunnus albacares</i>)				
Tuna Head waste	59.9±1.01	14.8±0.35	13.5±0.7	11.7± 0.61
Tuna Viscera	77.0±1.38	16.1±0.42	4.6±0.5	1.7±0.42
Tuna Dark Muscle (Raw)	74.0±0.76	21.5±0.31	2.87±0.30	1.5±0.19
Tuna Dark Muscle (Pre-cooked)	73.3±0.83	24.5±0.70	0.57±0.06	1.1±0.15
Fish head & viscera				
<i>Sardinella longiceps</i>	67.3±0.93	19.0±0.36	11.63±0.70	1.8±0.20
<i>Rastrelliger kanagurta</i>	71.3±0.93	20.7±0.56	6.7±0.57	1.4±0.13
<i>Nemipterus japonicus</i>	77.4±0.55	18.0±0.66	3.0±0.37	1.7±0.31
Cephalopod processing waste				
<i>Sepia pharonis</i>	89.0±0.57	9.6±0.42	0.47±0.08	0.9±0.20
<i>Loligo duvacelli</i>	91.7±1.57	7.7±0.58	0.34±0.06	0.38±0.04
Shrimp head waste				
<i>Metapenaeus dobsoni</i>	82.6±0.50	9.7±0.50	2±0.10	5.7±0.45
<i>Parapenaeopsis stylifera</i>	81.0±1.01	9.5±0.45	4.7±0.4	4.8±0.33
<i>Penaeus indicus</i>	79.7±0.66	17.2±0.32	0.70±0.025	1.84±0.27
<i>Aristus alcockii</i>	77.4±0.53	9.7±0.75	8.13±0.25	4.5±0.46

4.3.2 Amino acid profile

The amino acid profile (Table 4.4) of various seafood processing waste proved the presence of essential amino acids such as Isoleucine, Leucine, Lysine, Valine and Histidine and was at maximum in tuna processing waste compared to other seafood waste varieties. Studies of Ovissipour *et al.* (2012) also indicated the presence of well-balanced essential amino acid content in tuna wastes like viscera. The fin fish processing waste consisting of head and viscera of oil sardine, mackerel and Japanese sea bream also reported a balanced proportion of essential and other amino acids as evident from the studies conducted by Mathew (2005), Park *et al.* (2008) and Zynudheen *et al.* (2008). The study is also in tune with the observations of Manjusha (2011) who reported a fair amino acid composition in cuttlefish and squid processing waste. The study also agreed with the observations of Kandra *et al.* (2012) and Zhao *et al.* (2006) in ascertaining that the shrimp head waste was an excellent source of protein and amino acids.

Table 4.4: Amino acid profile (mg/100 g tissue)

	Aspartic acid	Glutamic acid	Serine	Glycine	Histidine	Arginine	Threonine	Alanine	Proline	Tyrosine	Valine	Methionine	Cysteine	Isoleucine	Leucine	Phenyl alanine	Lysine
Tuna (<i>Thunnus albacares</i>)																	
Tuna Head waste	7.16	14.13	2.89	3.16	3.69	6.19	3.66	4.75	1.83	1.56	1.01	2.29	4.63	7.08	12.78	4.18	9.16
Tuna Viscera	4.91	7.61	1.94	2.41	2.96	4.56	2.68	3.26	2.27	2.29	3.72	1.93	6.3	3.1	5.21	2.31	5.55
Tuna Dark Muscle (Raw)	8.12	16.18	4.3	15.15	62.9	1.75	4.43	41.56	3.1	2.52	6.98	1.85	8.1	3.97	7.7	2.8	38.1
Tuna Dark Muscle (Pre-cooked)	7.32	15.62	3.6	14.69	58.6	0.995	3.98	39.69	2.96	1.98	6.12	0.895	7.89	3.24	6.935	2.1	36.93
Fish head & viscera																	
<i>Sardinella longiceps</i>	8.17	14.33	3.91	3.77	5.57	6.47	4.66	4.88	1.93	3.41	4.73	2.8	1.63	3.29	5.97	3.93	10.64
<i>Rastrelliger kanagurta</i>	3.47	4.55	1.68	2.99	1.29	2.24	1.62	2.61	1.93	0.98	2.01	-	-	1.52	2.72	1.49	2.58
<i>Nemipterus japonicus</i>	0.634	1.087	0.396	0.532	-	0.615	0.442	0.473	0.335	0.332	0.481	0.344	-	0.529	0.987	0.586	0.94
Cuttlefish processing waste																	
<i>Sepia pharotis</i>	7.68	8.99	2.65	2.44	3.01	5.18	2.83	3.36	5.96	0.49	1.36	1.17	0.65	2.57	5.23	0.63	7.24
Squid processing waste																	
<i>Loligo duvacoeli</i>	8.9	7.9	6	3.82	1.7	5.6	5	4	5.1	4.9	4.3	3.51	1.2	4.5	7.9	4.2	8.8
Shrimp head waste																	
<i>Metapenaeus dobori</i>	8.15	13.09	3.28	6.09	2	6	3.94	1.24	5.95	2.15	1.96	1.62	1.85	2.04	4.52	3.2	11.71
<i>Parapenaeopsis stylifera</i>	17.07	11.84	5.5	8.66	2.49	6.43	5.46	7.07	4.55	1.68	4.28	2.05	1.46	4.86	10.45	4.28	9.97
<i>Penaeus indicus</i>	10.02	17.73	3.33	7.75	2.26	2.05	3.2	6.86	7.53	1.83	3.48	3.87	0.74	3.1	7.24	3.33	8.51
<i>Aristus alcockii</i>	9.24	16.88	3.05	5.89	3.27	5	3.03	5.56	2.69	3.39	3.23	2.1	0.95	4.9	7.34	2.34	8.07

4.3.3 Fatty acid profile

According to the fatty acid profile analysis of different seafood processing waste varieties performed, the cephalopod processing waste consisting of squid and cuttlefish waste reported the maximum concentration of polyunsaturated fatty acids (PUFA) followed by fish head and viscera. The observation was in sync with the studies conducted by Nair *et al.* (2011) and Rai *et al.* (2012). The study also inferred that the saturated fatty acids (SFA) was maximum in tuna processing waste while monounsaturated fatty Acids (MUFA) was highest in the shrimp processing waste, which was also highlighted by Panggatand Rivas (1997), Nguyen *et al.* (2011), Sánchez-Zapata *et al.* (2011) and Gopakumar and Nair (1975) in their studies. Fig. 4.2 depicts the percentage composition of MUFA, PUFA and SFA in seafood waste.

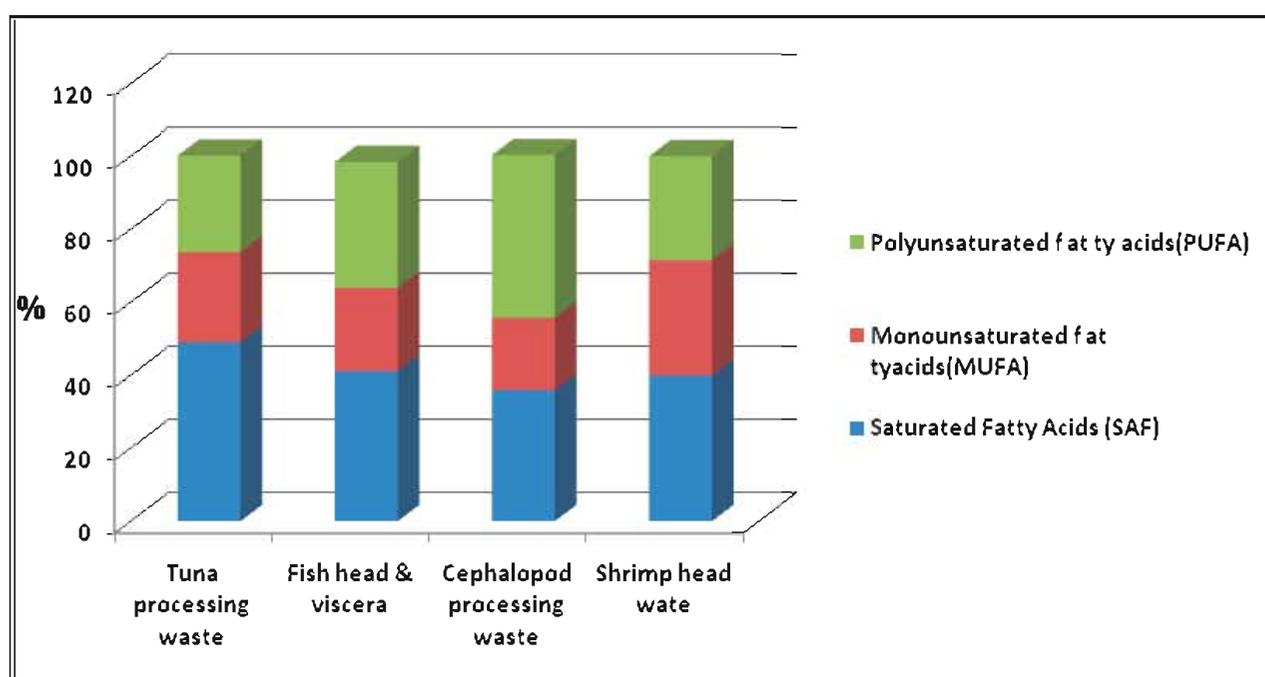


Fig. 4.2 Percentage composition of MUFA, PUFA and SFA in seafood waste

4.3.4 Chitin content

The analysis of chitin content of the shell waste from shrimp processing industry in the area showed that the range of chitin among them was within the range of 4.4 to 5.2 % as evident from Table 4.5, which is supported by the findings of Dutta *et al.* (2004).

Table 4.5: Chitin content in shell waste

Chitin content	%
<i>Metapenaeus dobsoni</i>	4.9 ± 0.51
<i>Parapenaeopsis stylifera</i>	5.2 ± 0.28
<i>Penaeus indicus</i>	5 ± 0.38
<i>Aristus alcockii</i>	4.4 ± 0.1

This confirms that shell waste is suitable for the production of shell waste derivatives.

4.4 Conclusion

- The proximate analysis of the seafood processing waste collected from the study area confirmed their superior nutritional quality with high content of protein and lipid .
- The amino acid analysis further proved the quality of the seafood waste protein, highlighting the presence of essential amino acids which in turn opens the possibility of developing protein rich by-products for human as well as livestock consumption.
- The fatty acid profile analysis highlighted the fact that the seafood waste materials are an excellent source of PUFA which could be industrially utilised for oil extraction of nutraceutical value.
- The chitin content of the shell waste available in the study area was also found to be within the acceptable range for industrial utilisation.



CHAPTER 5

DEVELOPMENT OF LIQUID PLANT FERTILIZER FROM SEAFOOD WASTE

Contents	5.1 Introduction
	5.2 Review of literature
	5.3 Materials and methods
	5.4 Results & Discussion
	5.5 Conclusion

5.1 Introduction

Subsequent to the qualitative and quantitative analysis of seafood solid waste from the study area (ASIB), it is evident that sufficient raw material is available there ready to be utilised for by product development. Disposal of such huge quantities of waste adds cost towards production for the producers. This also incurs management issues for the authorities and most often environmental problems for the local residents. The high nutritional quality of the seafood waste highlights the necessity of diverting the nutritional pool available in the form of waste to the food web for the benefit of food security of the country. In the case of shellfish waste generated in the study area, the utilization is fairly organized, for derivatives such as Chitin, Chitosan etc. The presence of three Chitin producing units in the area proves this fact. Other technologies such as extraction of pigments and protein from the shellfish waste are still at the research level or the technology is not economically feasible for the entrepreneurs. Similar is the case of other product options from seafood wastes such as Tuna oil, Enzymes, Protein supplement, Cuttlefish and squid ink etc. where the material is not sufficiently available in an uninterrupted supply to support an industry. Also the technology is expensive for small scale dissemination. The present study aims at identifying eco-friendly and low investment technology, which could be utilized for diverting the bulk quantities of seafood waste generated, without much investment and technological knowhow.

The entrepreneur should consider the following aspects before taking up such a task.

- Should be eco-friendly in practice.
- Should require minimum monetary input.
- Should not create any negative externalities to the community residing near the area of practice.

- Should be easy to understand and suitable for laymen to practice without much technical assistance.
- The expertise of the technology should be readily available in the area.
- The production of the product should not create additional chemical or organic waste.
- Should not be energy consuming.

Considering the above criteria, the technology identified to be suitable for dissemination in the study area was the production of silage using seafood industrial offal. The major product developed and utilized from fish silage throughout the world is animal feed. The production of animal feed from fish silage requires inputs such as rice/wheat bran and further drying, to be transformed into an end product. Moreover, the reduced demand of local made animal feed and competition from commercial branded feeds makes the option least preferred for waste utilization in the study area.

The recent awareness among the consumers about safety of vegetables and other agricultural produces, treated with chemicals as pesticides and fertilizers, paves a new possibility for utilizing seafood silage as an organic liquid fertilizer. The excellent fertilizer qualities of the seafood waste could be utilized for the development of a low cost and low investment liquid fertilizer for giving a boost for the budding organic agricultural movement. Other than this, the liquid seafood silage could be directly utilized for the purpose without much alteration to its physical state, thereby limiting financial inputs and ensuring better return. In this chapter, a general review is given on fish silage and foliar fertilization followed by the development of acid silage from three different seafood waste sources and converting them into liquid fertilizers. The key fertilizer qualities of the developed formulations are also analyzed in order to assess their potential as effective liquid foliar fertilizers.

5.2 Review of literature

5.2.1 Seafood silage

According to FAO (2003), fish silage is a “liquid product developed from the entire fish or parts of it, to which acids, enzymes or lactic-acid-producing bacteria are added, with the liquefaction of the mass aggravated by the action of enzymes from the fish”. Ockerman (1992) observed that fish silage is the product of the process of preserving and storing wet biological material with the help of acid in a silo. The production of fish silage was started in Sweden in 1936 to utilize fish waste for feeding purpose of livestock (Gopakumar, 1997). It is a stable liquid with a malty scent with all the moisture contained in the raw material (Raghunath and Gopakumar, 2002). The substrate for fish silage production are usually fish offal (head, bone, gut, fins, liver, roe), unsold whole fish (undersized, low-value, bycatch, spoilt), and shrimp processing wastes (Bertullo, 1984). Jayawardena *et al.* (1980) and Vanwyk and Heydenrych (1985) noted that fresh fish produced better silage than fish in which spoilage had set in, because of the risk from bacterial toxins, formation of biogenic amines and products of oxidative rancidity.

Fish bones and shrimp processing wastes are less favored for silage production since the buffering of the acid by calcium from fish bones and large deposits of undigested fractions of chitin formed, require large volumes of acid to neutralize (Trevino *et al.*, 1982). The conventional methods used for fish silage production are acid preservation and microbial fermentation. For both methods, the raw material has to be cut and crushed into pieces. Verburg and Freeman (1984) described a method that does not involve mincing of fish substrates. This silage produced varies between brown and grey viscous suspension, depending on the substrate and degree of autolysis (Disney and James, 1980).

5.2.2 Seafood silage production using acid ensilation method

Acid silage was developed in 1920 by A. I. Virtanen, using hydrochloric and sulphuric acid for the preservation of forages. Experiments with fish began in Sweden in 1936, using hydrochloric, sulphuric, and formic acids and sugars. The fish is partially digested and preserved by the acid (James, 1996). The most commonly used organic acids are propionic, acetic and formic acids (Hisano and de Pietro, 2013). Disney *et al.* (1978) recommended a set of precautions for the successful production of acid silage such as limiting the size of the raw material by 3-4 mm thorough dispersal of acid throughout the minced fish, to avoid air pockets of untreated material, to prevent bacterial spoilage, to bring about rapid liquefaction and to maintain the temperature above 20°C to prevent slowing down of liquefaction. For silage preparation from fish with a high mineral content, an 8% (v/w) of a 1:1 v/v mixture of formic and propionic acids may be essential (Ariyani and Buckle, 1991). The liquefied silage separates into 3-4 layers with an oily layer at the top, occasionally with an underlying emulsified layer, an aqueous middle layer which contains the liquefied proteins and a sediment layer containing the undigested protein, scales and bones (Raghunath and McCurdy, 1987). The pH below 4 attained through the addition of acids, prevent growth of bacteria and fungi, as well as kill fish parasites and their eggs. For whole fish, 3-4% formic acid is adequate (Arruda *et al.*, 2007), but the quantity depends on the amount of bones and scales present in the raw material which may neutralize the acid. Organic acids are comparatively expensive than the common inorganic acids but due to their weaker acidic nature, do not require neutralization before being administered as an animal feed ingredient (Tatterson, 1982). In addition, organic acids have fungicidal properties (Raa *et al.*, 1983).

5.2.3 Liquefaction in seafood silage

Fagbenro (1994) observed that the preserved fish gradually liquefies because of the endogenous proteases and lipases catalyzing the degradation of proteins into 28 peptides and amino acids and fats into free fatty acids, diglycerides, monoglycerides, and glycerol. According to Wheaton and Lawson (1985) the enzymes can originate from viscera and digestive organs, muscle tissue, plants, and microorganisms. Proteases from stomach, Intestine muscle and bacteria were found to contribute to the total proteolytic activity in fish silages (Gildberg and Raa, 1980). The enzymes largely responsible for liquefaction were those of the gut, skin and other

parts of the fish (Oulavallickal, 2010). Autolysis, indicated by liquefaction, is measured by a decline in viscosity of fish silage or by an increase in the volume of the aqueous phase on centrifugation. Liquefaction during silage production is usually completed within 7 days at 23-25°C (Raa and Gildberg, 1982). Liquefaction rate is influenced by the activity of digestive enzymes, proximate composition of raw materials, pH, temperature, and kind and percentage of preservative acids (Raa *et al.*, 1983). The undigested proteins are peptide aggregates held together by non-covalent forces (Hall *et al.*, 1985). It has also been reported that the autolysis of the silage does not complete with the presence of residue defiant to proteolysis.

5.2.4 Nutritional characteristics of seafood silage

The nutritional composition of fish silage is almost similar to fish except a slight increase in moisture content. The protein content of the silage is in the range of 16-19% (Rattagool *et al.*, 1977). Neethiselvan *et al.* (2001) also reported that the protein content of different types of fermented silages were more or less same as that of fresh fish. Sidwell (1981) indicated that the fat content of fish silage varies according to the fat content of the species used. Babu *et al.* (2005) reported a fat content of 3.6% to 5.1% in acid silage compared to 1% fat in fermented silage. The ash content of silage also depends on the nature of the raw material used and a higher level of ash can be expected if fish waste is used for ensilation. In general, acid silage contains less ash compared to fermented silage with the same raw material, possibly due to the minerals present in the molasses added. James *et al.* (1977) observed that free amino acids in both acid and fermented silages decreased slightly after storage for 6 months. The loss of tryptophan in stored fish silage is common, mainly at high temperature above 30°C (Haaland and Njaa, 1989). Disney *et al.* (1978) stated that histidine may also be limiting in fish silage prepared from partly spoiled fish and stored for long periods. It was also found that portions of free phenylalanine, arginine and glutamic acids were degraded with a loss of over 9% of the amino nitrogen as ammonia over a 6-week storage period without any degradation products of tyrosine (Stone and Hardy, 1986).

5.2.5 Foliar fertilization

Weinbaum and Neumann (1977) observed that the process of foliar penetration of a leaf, applied with a solution is quite complex and depends upon an array of environmental and plant factors. Kannan (2010) observed that foliar diffusion takes place by means of the cuticle, cuticular cracks, stomata, leaf hairs and other dedicated epidermal cells. Plant leaves are organs largely specialized in capturing light and CO₂, which play a critical role in plant survival and productivity (Sinha, 1999). Fernandez and Eichert (2005) state that the written evidence for the ability of plant leaves to absorb water from the environment dates back to the 17th century, and the absorption of mineral elements by leaves with the subsequent physiological effect in plants was first demonstrated in the 19th century.

The application of foliar nutrient sprays in agriculture is more and more widespread, since they are more environmentally friendly and target-oriented, as compared to root treatments (Oosterhuis, 2009). Marschner (1995) observed that in specific cases, foliar applications

of fertilizers can supply the plant with nutrients more rapidly than through root uptake. He also indicated the usage of foliar fertilizers in supplementing soil-applied nutrients, to compensate for decreased root activity, and to increase protein content of cereal seeds and calcium content in fruits. The efficiency of a nutrient spray is generally assessed in relation to its effect on yield and quality parameters (Nyomora *et al.*, 2000; Pestana *et al.*, 2001; Wojcik and Szwonek, 2002; Alcaraz-L'opez *et al.*, 2004; Dong *et al.*, 2005; Lester *et al.*, 2006; Singh *et al.*, 2007; Amiri *et al.*, 2008). Foliar spray field studies conducted in the past were chiefly carried out with fruit species such as grapevine, citrus, apple, peach etc and assessed the effect of various nutrient elements such as:- Phosphorous, Nitrogen, Iron, Magnesium, Boron, Zinc and Manganese. Johnson *et al.* (2001) emphasized the need to provide high concentrations to ensure the foliar uptake of macronutrients such as N and K, which are required by plants in greater amounts and are not likely to be met only by foliar treatments.

5.2.6 Foliar fertilization facts

Fernandez and Eichert (2009) observed that the foliar uptake studies carried out in the last 30 years investigated the mechanisms of cuticular dissemination. They indicated that in contrast to roots, the outer walls of epidermal cells of all aerial plants are covered by a hydrophobic cuticle, which is the limiting barrier for the transport of water and solutes and also that environmental factors such as relative humidity, light and temperature play a role concerning the penetration of a leaf applied solution. Barthlott and Neinhuis (1997) and Schonherr (2000), reported that the leaf water repellence is chiefly related to the epicuticular wax crystalloids which cover cuticular surfaces. Schonherr and Schreiber (2004) stated that the mechanism of cuticular penetration of water and ions is not fully understood but may occur due to the existence of aqueous pores. The upper as well as the lower leaf surfaces (Fig 5.1) are involved in the process of penetration of an applied solution and an increased penetrability of the lower epidermis versus the upper is noticed due to stomatal and cuticular variations

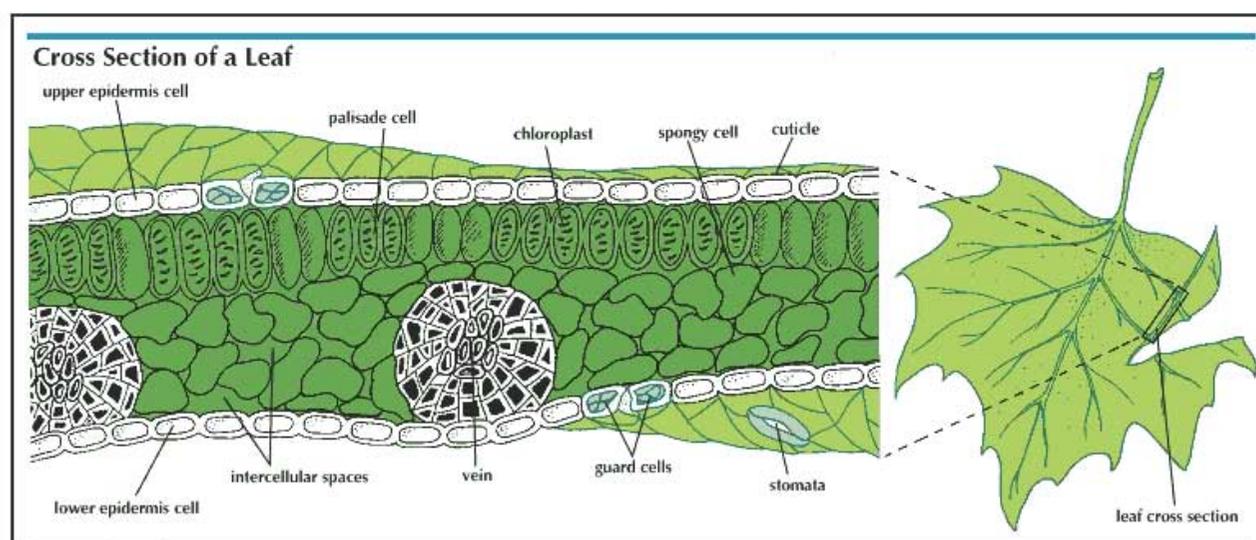


Fig. 5.1: Leaf cross section of a plant

(Source: <http://kids.britannica.com/comptons/art-53561/Cross-section-of-a-leaf>)

between both leaf sides. Structure and composition of the cuticle as well as the morphology, distribution and size of the stomata plays an important role in the penetration of foliar sprays. Numerous foliar application studies pointed the major role of light in stimulating foliar penetration through the abaxial leaf side (Outlaw, 2003). The spray-application process thus comprises of a sequence of complex interrelated activities such as, preparation of an active ingredient, atomization of the spray solution, transport to the target plant surface and droplet impaction, spreading and retention on the leaf surface, residue formation and penetration into the leaf (Brazee *et al.*, 2004).

5.2.7 Foliar Nutrition

It is observed that the three major elements, nitrogen, phosphorus, and potassium receive the highest place of importance among the necessary plant nutrient elements (Singh and Singh, 1973). Studies also prove that the application of these three major elements individually or in combinations has been made to improve the nutrient status, growth, flowering and fruiting, and the controlling of the nutrient deficiencies of different plants (Jack *et al.*, 2006). He also observed that the causes of low yield in plants could be due to inappropriate cultural operations. The inputs of N.P.K. fertilizers played a significant role in successful production of chillies. The foliar fertilizers are the best option for delivering the required nutrients to plants as they are known to immediately deliver nutrients to the tissues and organs. Baloch *et. al.* (2008) observed that 80 per cent of the phosphorus applied through conventional fertilizers get fixed up in the soil but up to 80 percent of the foliar-added phosphorus is directly absorbed. Islam *et.al.* (2003) stated that the micro-nutrients are utilized by plants in important life supporting processes or activities and are removed by plants from the soil and thus it becomes essential to replenish them with suitable balanced nutrient fertilizers to avoid nutrient deficiency. They also observed that balance between macro and micronutrients are very much important for plants to avoid antagonistic activities which may lead to poor growth and yield, affecting quality of leaf. Table 5.1 indicates the classification and characteristics of essential nutrients.

5.2.8 Seafood silage as a liquid fertilizer

It has been proven that the organic wastes contains compounds, which are capable of promoting plant growth (Day and Katterman, 1992), and seafood processing waste waters do not contain known toxic or carcinogenic materials unlike other types of municipal and industrial effluents (Afonso and Borquez, 2002b) which makes it suitable for fertilizer application. According to USDA, (2002), liquid fish products are considered synthetic plant or soil amendments because acid is often added to adjust the pH and the National Organic Program (NOP) allows the addition of organic or mineral acids for this purpose. But the amount of acid used cannot exceed the minimum needed to lower the pH to 3. Kim *et al.* (2010) also reported that only a few studies on reutilization of biodegraded waste products as liquid fertilizer are available.

Booth (1965) reported that the use of fish and seaweed products improved crop yield, seed germination, insect and fungal disease resistance, and low temperature tolerance. Fish

Table 5.1: Classification and characteristics of essential nutrients.

Nutrient	Characteristic
PRIMARY	
Nitrogen (N)	All converts to nitrate. Nitrate can leach from soil. Ammonium held to soil. Plants use mostly nitrate, some ammonium.
Phosphorous (P)	Easily tied up by soil and made unavailable to plants. Availability reduced at high or low pH and soil temperature below 50°F. Form for plant use varies with soil pH.
Potassium (K)	Increases size and quality of fruit.
SECONDARY	
Calcium (Ca)	Major component of cell wall. Does not move within plant. Deficiency related to blossom end rot of tomato and others. Can be deficient in acid soils, but corrected by liming.
Magnesium (Mg)	Deficiency may show on sandy, acid soils. Can be corrected with dolomitic limestone during pH adjustment.
Sulphur (S)	Can be deficient in acid soils.
MICRONUTRIENTS (TRACE ELEMENTS)	
Zinc (Zn)	Terminal growth areas affected first. Deficiency can be caused by excess phosphorous.
Iron (Fe)	Deficiency can be induced by high manganese at low pH. Usually associated with high pH soil or excess liming.
Copper (Cu)	Not usually deficient. May be linked to fruit cracking in tomato.

Adapted from McCraw and Motes (2007)

soluble nutrients (FSN) when applied as a soil drench fertilizer to greenhouse-grown plants, generated similar results in growth and yield, comparable to plants receiving an inorganic fertilizer (Aung and Flick, 1980; Emino, 1981). Soil fertilization with FSN also augmented the mineral content of the edible components of peas, tomatoes, lettuce, and radish which was above those same vegetables fertilized with standard nutrient solution (Aung *et al.*, 1983).

Abbasi *et al.* (2004) demonstrated that fish emulsion added to peat-based substrate or soil was effective in reducing incidence and severity of seedling damping-off diseases. They also found that foliar sprays of diluted solutions of fish emulsion can reduce bacterial spot severity in tomatoes and peppers and also increased the fruit yield, concluding that the fish emulsion not only acted as a disease managing product but was also found to be an excellent source of fertilizer. Prescott *et al.* (2000) reported that, while using fish silage treatments, the organic material was ground and ensiled in formic acid for weeks before application, therefore

most of the N in the silage was made available immediately after application. Fish silage has been reported to be very rich in most macronutrients and therefore a useful fertilizer for nutrient-deficient plantations (McDonald et al., 1994).

5.2.9 Application of natural pigments from seafood waste as a foliar color enhancer

Matsuno (2001) indicated that the crustaceans are rich in carotenoids. Shahidi *et al.* (1998) also pointed that the major carotenoids in shrimp are astaxanthin and its esters. Goodwin (1984) observed that the pigments may also exist bound to macromolecules such as proteins or chitin. Chayen *et al.* (2003) attributed the color of shell in crustaceans to combination of astaxanthin with proteins to form carotenoproteins. Manjabhat *et al.* (2006) assessed the quantitative and qualitative distribution of carotenoids in the body components of deep-sea shrimp *Solonocera indica* and *Aristeus alcocki*, from Indian waters. It was inferred in their study that the total carotenoid content was highest in *A. alcocki* cephalothoraxes (185.3 µg/g).

A study on direct effect of natural pigments in enhancing the color of plants and flowers has not been attempted yet. The coloration in flowers has been attributed to a range of factors such as nutrition, pH, temperature and most importantly type and amount of pigmentation. Carotenoids have been attributed for the yellow-red coloration in plants. As natural carotenoids are present in seafood waste such as shrimp waste, this study attempts to analyze the effect of foliar sprays developed from shrimp head waste rich in natural pigments on a red vegetable such as Amaranths. The shrimp head foliar preparations were applied with and without ensuring carotenoid stability through restricting air, temperature and light (Armenta and Guerrero-Legarreta, 2009). The tuna viscera and fin fish waste based foliar fertilizers were also analyzed for their effect on plants.

5.3 Materials and methods

5.3.1 Raw materials

The raw materials for silage production were carefully selected with an objective of getting an end product with maximum liquefaction and reduced wastage. Tuna viscera (TV) (*Thunnus albacares*) and finfish waste (FW) (*Nemipterus japonicus*) were found promising for the purpose due to the presence of high amount of digestive enzymes. Cephalothoraxes of deep sea shrimp (SH) (*Aristus alcockii*) were used for silage production for the purpose of analyzing the effect of natural pigments when applied as a liquid fertilizer.

5.3.2 Preparation of acid silage from seafood waste

The raw materials were freshly collected from the pre-processing units and immediately transferred to the lab under chilled conditions. The raw materials were then weighed and homogenized thoroughly in order to ensure uniform distribution of enzymes and acid during the treatment. Then 3.5% w/v 85% formic acid was added to TV and FW and thoroughly mixed and kept for aging in closed plastic containers with daily mixing for a period of 7 days.

In the case of deep sea shrimp cephalothoraxes, the acid silage was prepared in line with the concept of analyzing the effect of pigments as a foliar spray component. As the shrimp

head waste is rich in Calcium Carbonate (CaCo₃), a higher concentration of organic acid is required for ensilation (Chen et al., 1983) than for TV and FW. It was found that 7% w/v of formic acid is suitable for preparing SH after trials with different concentrations of acid. The shrimp head was mixed with TV in various concentrations of 25% (SH75:TV25), 50% (SH50:TV50) and 75% (SH25:TV75), in order to accelerate the enzymatic hydrolysis owing to the elevated levels of digestive enzymes in tuna viscera. The TV and SH wastes were homogenized and blended at aforesaid ratios and 7% w/v of 85% formic acid was added and thoroughly mixed.

For preserving the pigments, the SH silage was prepared in two lots. The Lot 1 was kept as control for ensilation under room temperature, with no antioxidant and light protection (NT). In Lot 2, SH was treated with 0.1% of butylated hydroxy anisole (BHA) (USFDA, 2011) as an antioxidant to prevent oxidation of pigments (T). The temperature was maintained below 25°C and the lot was kept in dark colored plastic bottles to prevent photo oxidation of pigments. Both the lots were kept for aging in closed plastic containers with daily mixing for a period of 7 days. All treatments were done in triplicates. Table 5.2 indicates the design of silage preparation from various seafood wastes.

Table 5.2: Seafood silage sample design for foliar spray preparation

Sl. No.	Foliar spray code	Composition	Treatment conditions and storage
1.	TV 100	100% Tuna Viscera	3.5% w/v, 85% formic acid, Room temp.
2.	FW 100	100% Fish Waste	3.5% w/v, 85% formic acid, Room temp.
3.	SH100 (NT)	100% Shrimp Head	7% w/v, 85% formic acid, Room temp.
4.	SH50:TV50 (NT)	50%SH + 50%TV	7% w/v, 85% formic acid, Room temp.
5.	SH75:TV25 (NT)	75% SH + 25% TV	7% w/v, 85% formic acid, Room temp.
6.	SH25:TV75 (NT)	25% SH + 75% TV	7% w/v, 85% formic acid, Room temp.
7.	SH100(T)	100% Shrimp Head	0.1% BHA, 7% w/v, 85% formic acid + 25°C Temp., Dark bottle.
8.	SH50:TV50(T)	50%SH + 50%TV	0.1% BHA, 7% w/v, 85% formic acid + 25°C Temp., Dark bottle.
9.	SH75:TV25(T)	75% SH + 25% TV	0.1% BHA, 7% w/v, 85% formic acid + 25°C Temp., Dark bottle.
10.	SH25:TV75(T)	25% SH + 75% TV	0.1% BHA, 7% w/v, 85% formic acid + 25°C Temp., Dark bottle.

5.3.3 Development of foliar fertilizer from seafood silage preparations

The liquefied TV and FW silages were later filtered through a muslin cloth to exclude particles and to get a fine liquid which could be used as a foliar fertilizer. The foliar fertilizer preparations (SH: TV) treated with BHA and stored under photo-temperature controlled conditions for pigment preservation were stored in a refrigerator (4-6°C). Table 5.3 indicates the foliar fertilizer yield from seafood waste silages.

Table 5.3: Foliar fertilizer yield from seafood waste silages

	Raw material (Silage)	Foliar fertilizer yield
1.	Tuna Viscera (TV)	80%
2.	Fish head & viscera (FW)	52%
3.	Shrimp head (SH100)	60%
4.	SH50:TV50	50%
5.	SH25:TV75	50%
6.	SH75:TV25	50%

5.3.4 Qualitative analysis of seafood foliar spray formulations

In order to assess the quality of various foliar fertilizer formulations developed from seafood waste resources, analysis of primary nutrients such as Nitrogen, Phosphorus, Potassium (N-P-K), secondary nutrients such as Calcium, Magnesium and Sulphur (Ca, Mg, S) and micronutrients such as Zinc, Iron, Manganese, Copper, Boron and Molybdenum (Zn, Fe, Cu, Mn, Cu, B, Mb) were conducted. The pH and the surface tension of the foliar formulations at different concentrations of spray were also assessed. The quality parameters of the foliar spray formulations were compared with a commercially available foliar fertilizer (COM). All analysis was done in triplicates.

5.3.4.1 N-P-K Analysis

5.3.4.1.1 Determination of Nitrogen (N)

1 gm of foliar spray liquid sample was accurately weighed into a digestion tube and the N was analysed according to Kjeldahl method (4.2.2.2).

5.3.4.1.2 Determination of Phosphorus (P)

Phosphorous was estimated according to the method of Rouser *et al.* (1970). 0.5 ml of foliar preparation was digested in test tubes by adding 10 ml of perchloric acid till colourless and then made up to 100ml using distilled water. 10 microliters of prepared solution was taken in a test tube and made up to 1ml and added 3.3ml water, 0.5 ml ammonium molybdate (2.5%), 0.5 ml ascorbic acid (10%), vortexed and kept in boiling water bath for 5 minutes and cooled. A blank and a standard (KH_2PO_4) were also taken simultaneously. The absorbance of the samples

was read spectrophotometrically (U-2800, Hitachi) at 800 nm against reagent blank and the values were expressed as percentage.

5.3.4.1.3 Determination of Potassium (K)

Potassium was determined using a BWB XP Flame Photometer (U.K.). Working standards in the range of 10, 20 and 40 ppm were made from the stock standard of 1000 ppm for K. The instrument was standardized with the above series of working standards. Samples were aspirated into flame and the corresponding readings were recorded. Calculations were carried out to find the concentration of K and the results were expressed as ppm.

5.3.4.2 Analysis of secondary and micronutrients

0.5 ml of foliar spray preparations were digested with 10 ml of nitric acid and perchloric acid mixture (9:4 ratio) in a test tube. The digested samples were made up to 100 ml for further analysis. The secondary nutrients such as Calcium, Magnesium and Sulphur (Ca, Mg, S) and micronutrients such as Zinc, Iron and Copper (Zn, Fe, Cu) were analyzed using the Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) instrument Optima 2000 DV Perkin Elmer. The calculation was made as follows:

$$\text{Mineral concentration in ppm} = \frac{\text{Instrument reading} \times \text{Make up volume}}{\text{Weight of the sample} \times \text{dilution factor}}$$

5.3.4.3 Analysis of foliar fertilizer pH

The pH of the foliar formulations in crude form and in 2% dilution was measured using the pH meter (Model: Cyberscan500).

5.3.4.4 Analysis of foliar fertilizer surface tension

The surface tension (ST) of the foliar spray formulations were analyzed using the Capillary rise method using the travelling microscope (WESWOX OPTIC India). The results were expressed as dynes/cm.

5.4 Results & Discussion

5.4.1 N-P-K Analysis

The N-P-K analysis of foliar formulations (Plate 5.1) in comparison with the commercial formulation (COM) showed that in the case of total Nitrogen (N), SH75:TV25 T & SH75:TV25 NT formulations topped the list followed by COM formulation, while the rest of the formulations showed a N% between 2-3%. For Phosphorus (P), TV100 reported the maximum value followed by FW100. The rest of the formulations showed a P % between 0.2-0.4 while the COM indicated a 0.28% of P. For Potassium (K) also the seafood foliar formulation, SH75:TV25 (T&NT) topped while the COM reported the second highest value of K. The rest of the formulations reported K% in a range of 4-5. Gaskell and Smith (2007) observed N as the most limiting nutrient to efficient and profitable vegetable production. Jagadeeswaran et al. (2005) also reported that the key nutrients necessary for plant development are N, P and K.

Table 5.4 indicates the results of NPK analysis and the NPK ratio of the foliar formulations.

Table 5.4: NPK analysis of seafood foliar formulations

Sl. No.	Foliar formulation	Primary nutrients (%)			NPK Ratio
		N	P	K	
1	TV 100	2.5 ± 0.10	10.6 ± 0.35	0.43 ± 0.004	2.5:10.6:0.43
2	FW100	2.6 ± 0.43	5.8 ± 0.35	0.23 ± 0.030	2.6:5.8:0.23
3	SH100 (NT)	2.6 ± 0.06	0.4 ± 0.04	2.04 ± 0.030	2.6:0.4:2.04
4	SH50:TV50 (NT)	2.5 ± 0.04	0.3 ± 0.04	4.85 ± 0.036	2.5:0.3:4.85
5	SH75:TV25 (NT)	8.2 ± 0.04	0.3 ± 0.04	7.37 ± 0.252	8.2:0.3:7.37
6	SH25:TV75 (NT)	2.9 ± 0.05	0.3 ± 0.03	2.65 ± 0.040	2.9:0.3:2.65
7	SH100 (T)	2.7 ± 0.04	0.3 ± 0.02	2.22 ± 0.040	2.7:0.3:2.22
8	SH50:TV50 (T)	2.3 ± 0.04	0.3 ± 0.05	4.72 ± 0.330	2.3:0.3:4.72
9	SH75:TV25 (T)	8.2 ± 0.03	0.4 ± 0.05	7.94 ± 0.140	8.2:0.4:7.94
10	SH25:TV75 (T)	2.8 ± 0.04	0.3 ± 0.03	2.44 ± 0.045	2.8:0.3:2.44
11	COM	4.4 ± 0.05	0.3 ± 0.04	7.63 ± 0.035	4.4:0.3:7.36

Analysis was done in triplicates

In the present study, when compared to commercial foliar spray, TV and FW formulations were low in potassium and nitrogen content, but very rich in phosphorus. The foliar formulations prepared from 100% shrimp head waste (SH) and their combinations with TV showed a comparable ratio with the COM formulation. Fig 5.2 shows the ratio of NPK among different test formulations in comparison with COM foliar spray which shows that while the TV and FW showed a P dominated ratio, SH:TV formulations especially SH75:TV 25 (T&NT) showed a balanced ratio. Studies also indicate that the insufficient supply of any of these nutrients during crop development will have harmful impact on the reproductive potential, growth and yield of the plant (Agbaje and Akinlosotu, 2004). Studies also show that foliar applications

of phosphorus and other macronutrients as a supplementary fertilizer are highly effective (Silberbush, 2002 and Pimple *et al.*, 2006). Grigg (1999) inferred that some nutrients like phosphorus becomes readily fixed in the soil and are often difficult for roots to absorb and hence it is better to be applied as foliar fertilizer. The utilization of seafood waste as a liquid fertilizer has been studied by many scholars (Gagnon and Berrouard, 1994; Blatt and McRae, 1998) and found to be an effective means for transferring NPK nutrients to plants.

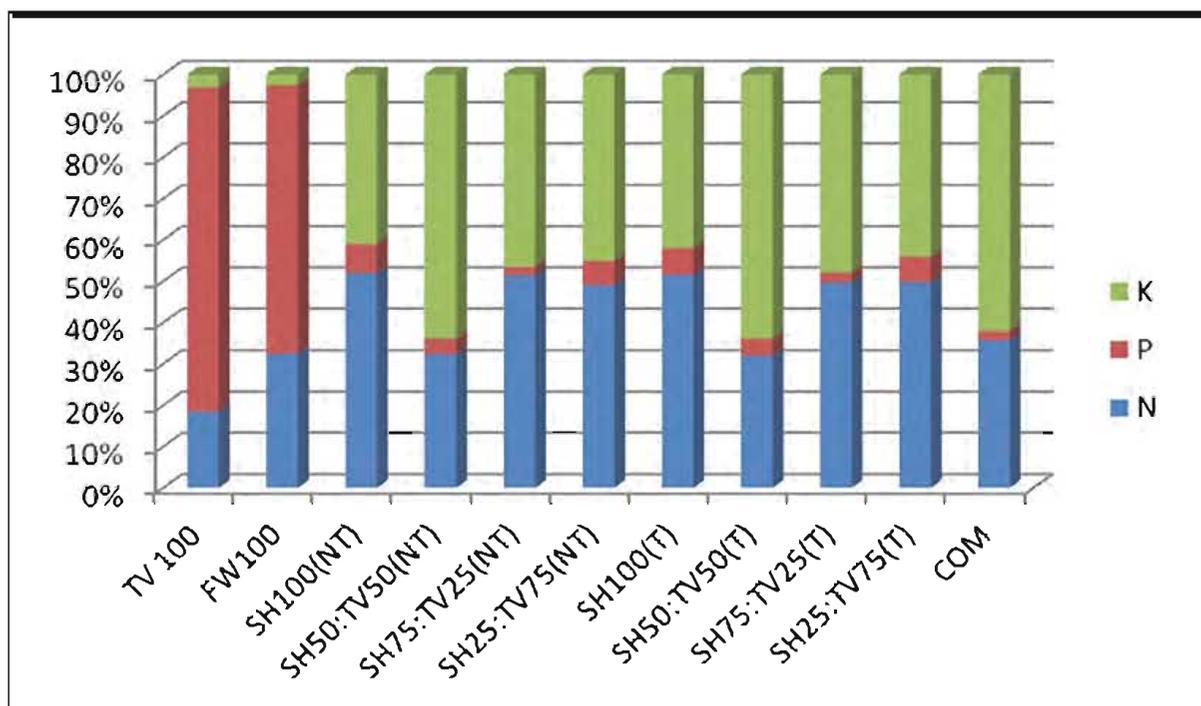


Fig. 5.2: NPK ratio of seafood foliar formulations in comparison with commercial foliar spray

5.4.2 Analysis of secondary and micronutrients

In the case of secondary nutrient analysis (Table 5.5), SH100 (T) and SH100 (NT) showed a maximum in Ca concentration with 2.204 and 1.8 ppm and the rest of the formulations fluctuated between 0.3-1 ppm. The COM reported a Ca concentration of only 0.114 ppm.

In the present study, the Mg concentrations were maximum in TV100 (0.2032 ppm) and FW100 (0.3096 ppm). The rest of the formulations reported readings between 0.07 - 0.1 ppm including COM which indicated 0.0756 ppm of Mg. The S concentrations were high in COM with 5.114 ppm, while for all of the seafood foliar formulations, the values were between 0.1-0.4 ppm. The micronutrients such as Zn, Fe and Cu were reported in maximum concentrations in COM of 0.0372, 0.1 and 0.023 ppm respectively. The study proves that in comparison to the COM formulation, the seafood silage based foliar formulations are more dependable as secondary and micronutrient source especially in the case of Ca, Mg and Fe. The COM formulation showed a micro nutrient ratio dominated by S, while the seafood formulations like TV showed a more balanced ratio of micro nutrients when compared to COM (Fig 5.3). The importance of secondary and micronutrients on the health of plants has been highlighted by many researchers (Hochmuth and Hanlon, 2000, and Fageria *et al.*, 2002). The foliar application of such nutrients

were also studied and recommended for plant fertilization (Durstberger *et al.*, 2008 and Vitosh *et al.*, 1994).

Table 5.5: Secondary and micronutrients in foliar formulations.

Sl. No.	Foliar formulation	Ca	Mg	S	Zn	Fe	Cu
1	TV 100	0.32 ± 0.004	0.2029 ± 0.0003	0.2032 ± 0.007	0.2035 ± 0.0004	0.094 ± 0.0040	0.20 ± 0.0005
2	FW100	1.13 ± 0.003	0.3091 ± 0.0004	0.3096 ± 0.005	0.3099 ± 0.0005	0.050 ± 0.0005	0.31 ± 0.0003
3	SH100(NT)	1.50 ± 0.26	0.1264 ± 0.0004	0.1268 ± 0.004	0.1271 ± 0.0040	0.019 ± 0.0004	0.13 ± 0.0004
4	SH50:TV50(NT)	0.54 ± 0.004	0.068 ± 0.0025	0.071 ± 0.004	0.073 ± 0.0005	0.043 ± 0.0040	0.07 ± 0.0015
5	SH75:TV25(NT)	1.45 ± 0.045	0.1069 ± 0.0003	0.1072 ± 0.005	0.1075 ± 0.0004	0.059 ± 0.0004	0.11 ± 0.0004
6	SH25:TV75(NT)	0.97 ± 0.004	0.0809 ± 0.0004	0.0812 ± 0.004	0.0816 ± 0.0004	0.037 ± 0.0004	0.08 ± 0.0026
7	SH100(T)	2.20 ± 0.004	0.1421 ± 0.0004	0.1426 ± 0.004	0.1429 ± 0.0005	0.036 ± 0.0004	0.14 ± 0.0004
8	SH50:TV50(T)	1.09 ± 0.004	0.1004 ± 0.0003	0.1006 ± 0.003	0.1009 ± 0.0004	0.039 ± 0.0004	0.10 ± 0.0003
9	SH75:TV25(T)	0.99 ± 0.004	0.0912 ± 0.0004	0.0916 ± 2.000	0.0919 ± 0.0003	0.064 ± 0.0005	0.09 ± 0.0004
10	SH25:TV75(T)	0.71 ± 0.004	0.12 ± 0.0045	0.125 ± 0.004	0.129 ± 0.0003	0.042 ± 0.0005	0.12 ± 0.0003
11	COM	0.11 ± 0.003	0.0754 ± 0.0004	0.0758 ± 0.004	0.0761 ± 0.0003	0.101 ± 0.0003	0.08 ± 0.0035

Analysis was done in triplicates

5.4.3 Analysis of foliar fertilizer surface tension

The capillary method analysis of Surface Tension (ST) of various seafood foliar formulations (Table 5.6) indicated that the ST of seafood foliar formulations diluted to ready to spray preparation of 1% were considerably lower (0.021 to 0.026) than the control of freshwater (0.065) and also similar commercial foliar spray (0.041) dilutions indicating their ability in spreading immediately after the application in a faster rate, increasing the chance of absorption rate through the leaf surface. Studies have shown that the surface tension of the applied liquid greatly influences spray retention and leaf wettability (Fernandez and Eichert, 2009; Murtic *et al.*, 2012).

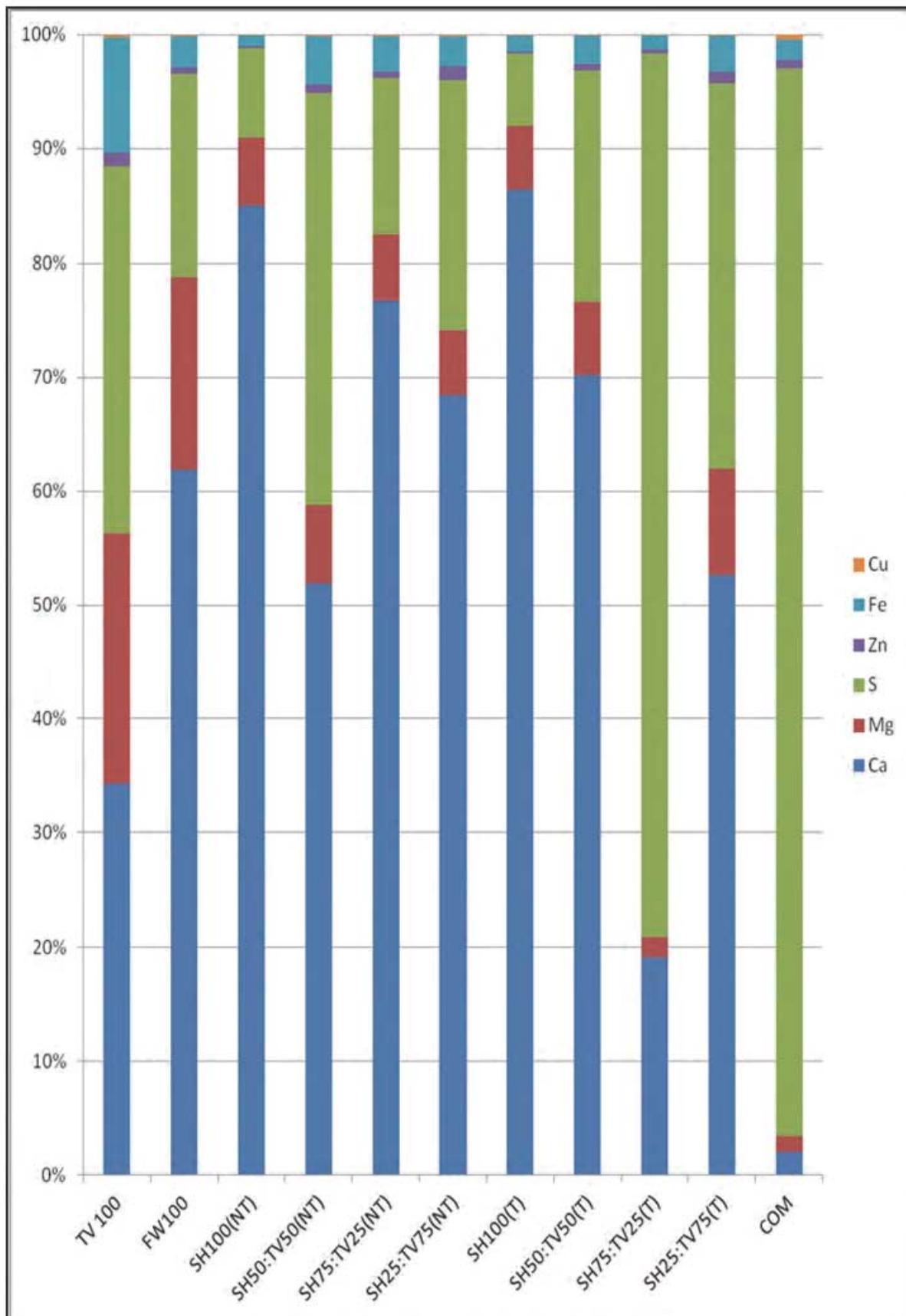


Fig. 5.3: Ratio of micronutrients in foliar formulations

Table 5.6: Surface tension of foliar formulations at 1% concentration

	ST (dynes/cm.)
Water (CNT)	0.065 ± 0.0004
COM 1%	0.041 ± 0.0003
TV 1%	0.026 ± 0.0031
FW 1%	0.021 ± 0.0004
SH100T 1%	0.023 ± 0.0005
SH100NT 1%	0.026 ± 0.0005
SH:TV(50/50) T 1%	0.023 ± 0.0003
SH:TV(50/50) NT 1%	0.025 ± 0.0003
SH:TV(25/75) T 1%	0.023 ± 0.0004
SH:TV(25/75) NT 1%	0.021 ± 0.0004
SH:TV(75/25) T 1%	0.023 ± 0.0004
SH:TV(75/25) NT 1%	0.023 ± 0.0004

Analysis were done in triplicates

Schonherr (2000) observed that close contact between an aqueous solution and the leaf surface can be achieved by the addition of suitable surfactants in the spray formulation (Fig 5.4). Stock and Holloway (1993) described surfactant composition and concentration, as key factors influencing leaf penetration of agrochemicals. The lower ST values of seafood based foliar formulations when compared to COM and water samples ensure higher nutrient penetration rates.



Fig.5.4: Effect of surfactants on surface tension
 (Source: <http://img.tradeindia.com/ffp/1/001/530/428.jpg>)

5.4.4 Analysis of foliar fertilizer pH

pH of the seafood foliar fertilizer formulations were compared with controls of tap water (CNT TW), distilled water (CNT DW) and commercial preparation in 1% dilution (Table 5.7). The seafood based foliar fertilizer preparations showed pH in acidic range (2.85 to 3.65) complementing the commercial foliar product (2.61).

Table 5.7: pH of stock and diluted foliar formulations

	Stock	1% dilution
COM	1.55 ± 0.05	2.61 ± 0.03
TV	3.34 ± 0.03	3.50 ± 0.04
FW	3.50 ± 0.40	3.65 ± 0.04
SH100T	3.04 ± 0.03	3.16 ± 0.03
SH100NT	2.94 ± 0.05	3.40 ± 0.04
SH:TV(50/50) T	2.92 ± 0.04	3.05 ± 0.04
SH:TV(50/50) NT	2.80 ± 0.40	3.04 ± 0.04
SH:TV(25/75) T	2.76 ± 0.05	3.04 ± 0.14
SH:TV(25/75) NT	2.67 ± 0.35	3.05 ± 0.04
SH:TV(75/25) T	2.51 ± 0.02	2.85 ± 0.04
SH:TV(75/25) NT	2.40 ± 0.04	2.87 ± 0.04
CNT (DW)	4.93 ± 0.45	NA
CNT (TW)	6.13 ± 0.04	NA

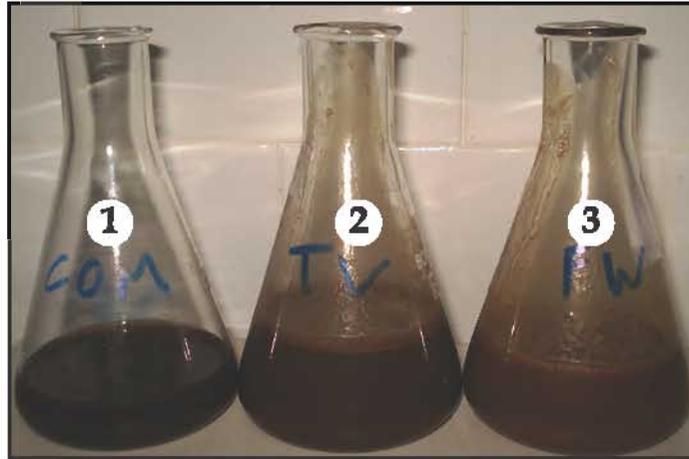
Analysis was done in triplicates

Wojcik (2004) has observed that the hydrogen ion concentration of solutions applied to the foliage of plants is an important factor in absorption and maximum absorption of nutrients occur at an acidic pH level. The maximum absorption of foliar urea, iron, phosphorus and sodium were exhibited by leaves when the solution pH was maintained low (El-Otmani *et al.*, 2000; Fernandez *et al.*, 2006; Fernandez and Ebert, 2005). The study establishes the suitability of seafood based foliar formulations as a plant agricultural input because when compared to commercially available foliar spray solutions, they exhibited a complementing range of pH.

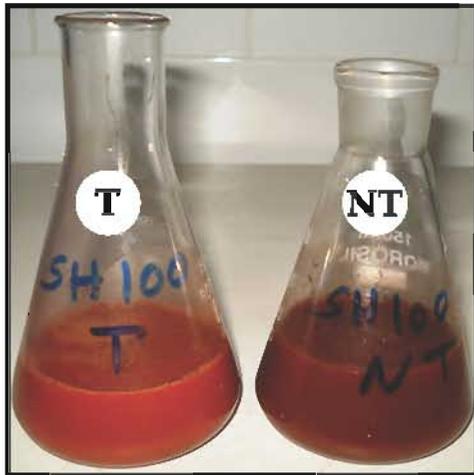
5.5 Conclusion

- The NPK analysis of the seafood silage based foliar preparations demonstrated that the primary nutrients such as Nitrogen, Phosphorus and Potassium were present in comparable ratios to that of commercial formulation used in the study.
- The secondary and micronutrient evaluation also revealed that the seafood foliar formulations were comparable to commercial sample and further more, some of them such as TV100 showed a more balanced ratio than the commercial sample.
- The analysis of Surface Tension (ST) of the foliar formulations at 2% level indicated that compared to commercial formulation and control (water), the seafood formulations demonstrated a low ST, favouring their nutrient absorption rate.
- The pH of the test formulations were also found to be in the desired range for maximum nutrient absorption confirming their candidature as potential foliar spray formulations.

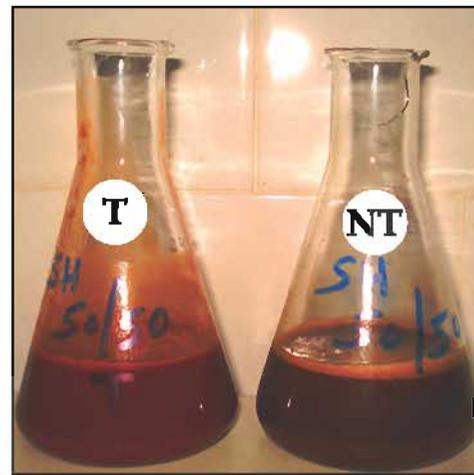




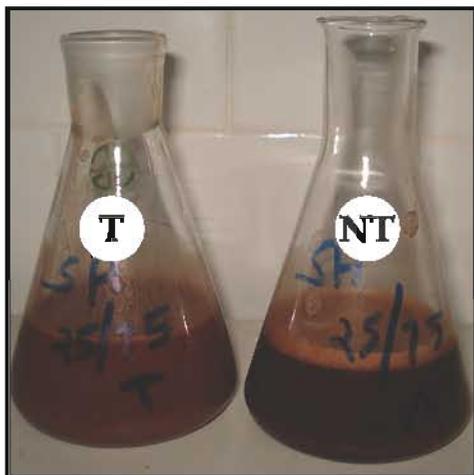
5.1(a): COM (1), TV100 (2) and FW100 (3) foliar formulations.



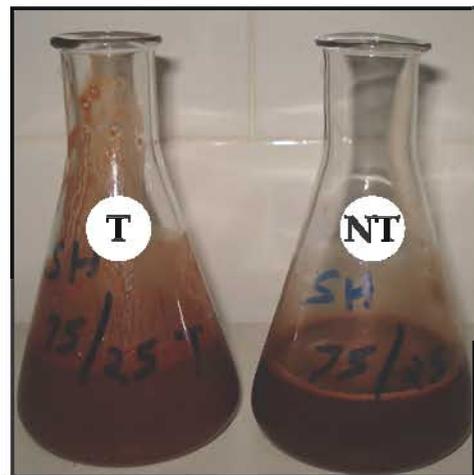
5.1(b): SH100 treated and non-treated formulations



5.1(c): SH50:TV50 treated and non-treated formulations



5.1(d): SH25:TV75 treated and non-treated formulations



5.1(e): SH75:TV25 treated and non-treated formulations

Plate 5.1: Different foliar formulations prepared from seafood waste

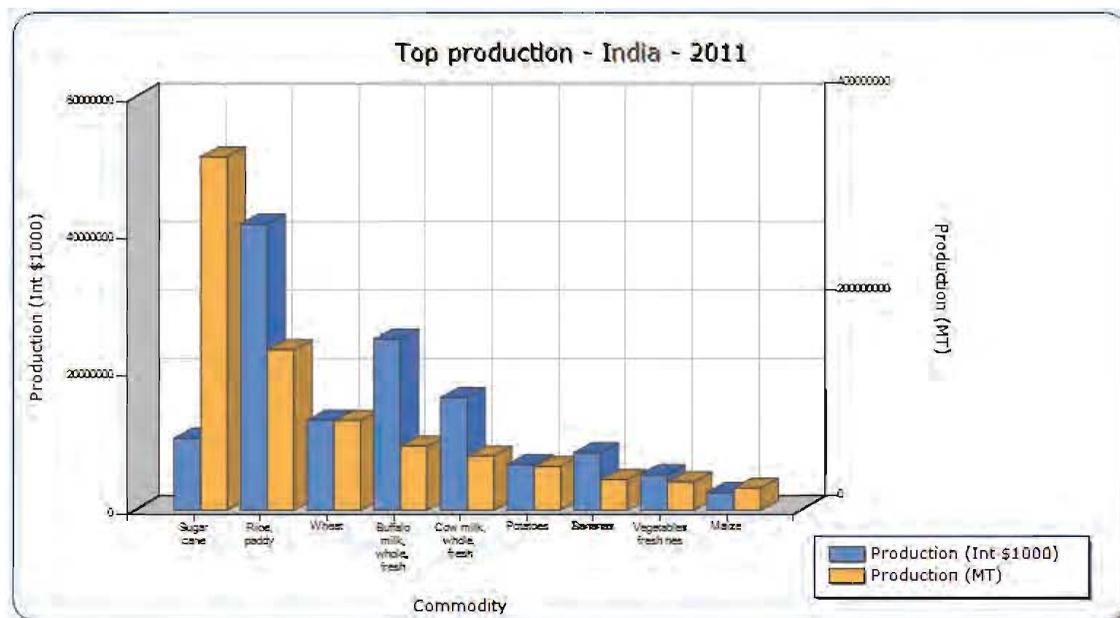
CHAPTER 6

EFFECT OF SEAFOOD SILAGE BASED FOLIAR FORMULATIONS ON BIOMETRICAL AND YIELD CHARACTERISTICS OF OKRA (*Abelmoschus esculentus*) AND RED AMARANTH (*Amaranthus tricolor* L.)

Contents	6.1 Introduction
	6.2 Indian Agriculture: Problems & Prospects
	6.3 Organic farming as a tool for agricultural sustainability
	6.4 Importance of organic fertilization and scope of seafood waste silage
	6.5 Section A Study on the effect of seafood silage based foliar formulations on the biometrical and yield characteristics of Okra (<i>Abelmoschus esculentus</i>)
	6.6 Section B Study on the effect of seafood silage based foliar formulations on biometrical and yield characteristics of Red Amaranth (<i>Amaranthus tricolor</i> L.)
	6.7 Conclusion

6.1 Introduction

Agriculture is considered as the backbone of Indian economy. The sector reported a total production of 829.57 million metric tonnes during 2010 (FAO 2010). Fig 6.1 depicts the total commodity wise agricultural production status of India in terms of value and volume.



Source: FAO, 2011

Fig. 6.1: Agricultural production in India during 2010.

6.2 Indian Agriculture: Problems & Prospects

According to Department of Agriculture & Cooperation, Ministry of Agriculture Government of India report on agricultural statistics (2011), the contribution of agricultural sector to the overall GDP of the country is reported to be reduced from 30 percent in 1990-91 to less than 15 percent in 2011-12. The report also states that the state of Indian agriculture is representing a shift from the conventional agrarian economy towards a service dominated one. This decrease has not been accompanied by a matching reduction in the share of agriculture in employment since about 52% of the total workforce is still employed by the farm sector. The study also raises the alarm on the crippling growth rate of the sector.

The reduction in growth rate of a sector, on which more than 50 percentage of the total workforce of a country is dependent, is definitely an alarming situation. The fact that, the sector concerned is a primary production sector raises the severity of the situation furthermore. Ryu (2011) stated that even though the green revolution played an important role in multiplying the production of Indian agricultural crops, it failed to account for the destruction of environment and lack of sustainability of its practices. Zwerdling (2009a) stated that many of the Indian agricultural experts find agriculture to be unsustainable and unprofitable, and some predict a rapid decline in agricultural output in future. Studies have also made observations that the excess usage of chemical fertilizers and pesticides has done considerable damage to the environment by heavily polluting the land, which has also resulted in exceptionally high rates of cancer among Indian farming families and also health problems on consumption of such produce (Zwerdling, 2009b).

Many researchers feel that a holistic way of approach is needed to tackle the crisis in Indian agricultural sector. It has been pointed out that unlike our present approach of agricultural

development through increase in agricultural area and yield, our future growth will need to increase yield with sustainable and improved management of shrinking land and freshwater resources (Kumar *et al.*, 2008). Murthy *et al.* (2009) observed that agricultural productivity and output can be increased sustainably through informed and locally adapted use of high-yield variety seeds and pest, disease, and nutrient management technologies.

6.3 Organic farming as a tool for agricultural sustainability

The organic way of farming is being projected as a possible and practical solution for reviving the farming sector of India to a sustainable level. Rajalakshmy (2011) observed that the organic farming has been a fundamental part of Indian agricultural practices since ancient times, till the use of chemical fertilizers became crucial under the Green Revolution program. She also stated that the domestic and the international market for organic products have multiplied through recent years with a 25 fold increase in the industry of organic products. India ranks 33rd in terms of the total area under organic cultivation and the country's export of organic products are expected to cross ₹ 2500 crores by 2012 (Bhattacharya and Chakraborty, 2005). According to Ramesh *et al.* (2010) the growing awareness, increasing market demand, increasing inclination of farmers to go organic and growing institutional support have resulted in more than 200% growth in certified area during the last two years. Organic farming offers an alternative, that is eco-friendly and healthy. But it also faces challenges as a budding sector with regard to financial viability, availability of organic inputs, and teaching of skills required for productive organic farming (Ryu, 2011).

6.4 Importance of organic fertilization and scope of seafood waste silage

Even though there is no dispute regarding the sustainability of organic farming, availability of suitable fertilizer with acceptable standard and sufficient quantity is one of the major concern. It is still not possible for small farmers to afford the cost and hence, plant nutrients must be sufficiently supplemented through compost and bio fertilizers, to bring in sustainability of production. Thus adequate awareness is required to augment soil with as much organic matter as possible (Saqib and Kaushik, 2001). According to FAO statistics (2005) the estimated annual available nutrient (NPK) contribution through organic sources is about 5 million tonnes, which is required to be increased, emphasizing that the organic manures have a significant role to play in nutrient supply. The report also stated that in addition to improving soil physico-chemical properties, the supplementary and complementary use of organic manure also improved the efficiency of mineral fertilizers. The Ministry of Agriculture, Govt. of India has also taken steps to promote organic farming through various schemes such as the National Project on Organic Farming (NPOF), National Horticulture Mission (NHM) and Rashtriya Krishi Vikas Yojana (RKVY) which still is going to increase the demand for organic fertilizers.

Application of organic extracts as growth stimulants in horticultural crops are fast gaining importance (Tarun and Kose, 2004). The foliar spray is observed to control pests and enhance the yield (Praveen *et al.*, 2007). Brown (2007) observed that in order to address the issue

of transient nutritional deficiencies occurring as a result of limitations in uptake or restrictions in nutrient delivery during periods of peak nutrient demand, many horticulturists are utilizing foliar fertilizers. Foliar fertilizers allow for highly localized and specifically customized nutrient applications that are lacking in solid or blended products, particularly for micronutrients.

Considering the qualitative and quantitative availability of primary, secondary and tertiary nutrients in silage prepared from seafood waste as explained in previous chapters, it could be inferred that it is a suitable candidate as a plant fertilizer. And also the fact that the only ingredient contained in seafood waste silage is the formic acid, which is a permitted organic input, (Yadav, 2010) make it suitable for organic fertilization. Vegetable farming is important in Indian Agricultural sector with a very high potential for organic adoption. Taking these factors into consideration, experiments were conducted to demonstrate the effects of seafood silage based liquid fertilizers on two common vegetables, Okra (*Abelmoschus esculentus*) and red Amaranth (*Amaranthus tricolor L.*), in comparison with a standard chemical fertilizer and a commercial foliar spray. These vegetables were selected for the study considering the local availability of seed, short growth duration and simple propagation procedures. The experiments were repeated twice for rectifying the errors and then the final experiment was conducted.

6.5 Section A

Study on the effect of seafood silage based foliar formulations on the biometrical and yield characteristics of Okra (*Abelmoschus esculentus*)

6.5.1 Introduction

Okra, *Abelmoschus esculentus L.* (Moench), is an agriculturally and economically important vegetable crop grown in tropical and sub-tropical regions of the world. Okra is suitable for cultivation as a garden crop as well as on large commercial farms and it is grown commercially in India, Turkey, Iran, Western Africa, Yugoslavia, Bangladesh, Afghanistan, Pakistan, Burma, Japan, Malaysia, Brazil, Ghana, Ethiopia, Cyprus and the Southern United States. India ranks first in the world and contributes 70% of the total world production with 4.5 million tonnes (FAO, 2010). Okra is cultivated for its green non-fibrous fruits or pods containing round seeds and the fruits are harvested when immature and eaten as a vegetable. The roots and stems of okra are used for clarification of sugarcane juice from which gur or brown sugar is prepared (Chauhan, 1972). Okra provides an important source of vitamins, and minerals .

6.5.2 About Okra

The foliar mode of applying fertilizers to the vegetable crops has been considered a valuable addition to the application of nutrients to soil system (Fageria *et al.*, 2009). Foliar application of nutrients on okra was studied by many scholars. The study conducted by Abbasi *et al.* (2010) proved the effect of foliar fertilization on okra when they compared three foliar fertilizer products individually, and integrated with soil fertilizer application at recommended nitrogen and phosphorus. The study endorsed the benefits of foliar fertilization by demonstrating the

enhanced growth traits of okra plants, viz. days to flowering, plant height, number of branches, number of fruits and fruit length, with the enhanced crop yield. Alkaff and Hassan (2003) reported better growth and yield qualities of okra in relation to foliar fertilization. The study outcome of Paliwal *et al.* (1999) elucidated maximum number of branches in okra plants in response to foliar fertilization. Many other studies also indicated the benefits of foliar fertilization in improving growth, yield, nutrient uptake and product quality (Naruka and Singh, 1998; Tumbare *et al.*, 1999; Naruka *et al.*, 2000; Chattopadhyay *et al.*, 2003).

6.5.3 Materials & Methods.

Field experiment was conducted to study the effect of seafood silage based liquid foliar formulations on the okra *cv. Arka Anamika*. The details of the materials used and methods adopted during the course of investigation are furnished in this section.

6.5.3.1 General Description

6.5.3.1.1 Location of experimental site

The field experiment was conducted in the experimental field located at School of Industrial Fisheries, lake side campus, Cochin University of Science & Technology, Cochin, Kerala during December 2009 – February 2010. The experimental field is situated at 9°57'51,90" N latitude and 76°16'58.58" E. longitude with an altitude of 15 ft above mean sea level and it is situated in the southern transitional belt of Kerala.

6.5.3.1.2 Climatic and weather conditions

The monthly meteorological data during December 2009 – February 2010 was recorded from the Meteorological Observatory of Indian Meteorological Department, Trivandrum Regional Branch, and is given in Table 6.1. The mean temperature ranged from 23.3°C (January) - 32.96 (February) and the mean monthly relative humidity ranged from 73.2 (January) to 80.2 (December). The entire cropping period received 43 mm rainfall from December to February. The highest maximum temperature recorded was 32.96°C (February) and the minimum temperature was 23.3°C (January). The lowest maximum and minimum temperatures recorded during the experiment were 32.4°C (January) and 23.3°C (January) respectively and maximum and minimum relative humidity were 80.2 per cent (December) and 73.2 per cent (January), respectively.

Table 6.1: Monthly meteorological data for December 2009 – February 2010

Sl. No.	Months	Temperature (°C)		Relative Humidity (%)	Mean Rainfall (mm)
		Mean minimum	Mean maximum		
1.	December 2009	24.13	32.7	80.2	0.23
2.	January 2010	23.3	32.4	73.2	1.2
3.	February 2010	24.7	32.96	78.2	0

6.5.3.1.3 Soil characteristics

The soil used for the experiment was medium deep brown. A composite soil sample was taken from the experimental area and its physical and chemical properties were analyzed, and the values obtained are furnished in Table 6.2.

Table 6.2: Physico- Chemical properties of soil

Component	Value	Reference
A. Physical properties		
Sand (%)	32.7±1.01	Pipette method (Day, 1965)
Clay (%)	9.5±1.38	Pipette method (Day, 1965)
Silt (%)	57.8±0.76	Pipette method (Day, 1965)
B. Chemical properties		
Total N (kg/ha)	265±1.7	Kjeldahl method (Subbaiah and Asija, 1956)
P (mg/kg soil)	71.6±1.57	Olsen's method (Jackson, 1967)
K (mg/kg soil)	84±1.67	Flame photometric method (Jackson, 1967)
Organic carbon %	1.87±0.83	Wet oxidation method (Jackson, 1967)
pH	6.75±1.8	pH meter (Piper, 1966)

6.5.3.2 Materials used

6.5.3.2.1 Seed

The okra *cv. Arka Anamika* seeds were purchased from the Central Nursery of Kerala Agricultural University, Thrissur. According to Praveen *et al.* (2007) it is a variety from inter-species cross between *Abelmoschus esculentus* and a wild species *Abelmoschus manihot* spp. tetraphyllum. It was released in 1984 by Indian Institute of Horticultural Research, Bangalore. Biometrically, plant is medium tall and grows to a height of 100 cm with short internodal length, less branched and with 5 splashes of purple pigmentation present on stem, petiole and lower surface of leaf. Leaves are green, small and deeply lobed; stem, petiole and leaves are hairy. It is resistant to yellow vein mosaic disease. It takes about 50 days for flower initiation, early maturing. The first picking of green fruits can be made within 58-60 days of planting. Fruits are medium, green, rough, five ridged and starts after 5th-6th node. It is a high yielding variety and gives about 115 quintal per ha of green fruits. During 1990, it has been identified for general cultivation under the All India Co-ordinated Vegetative Improvement Project (Sharma and Arora, 1993).

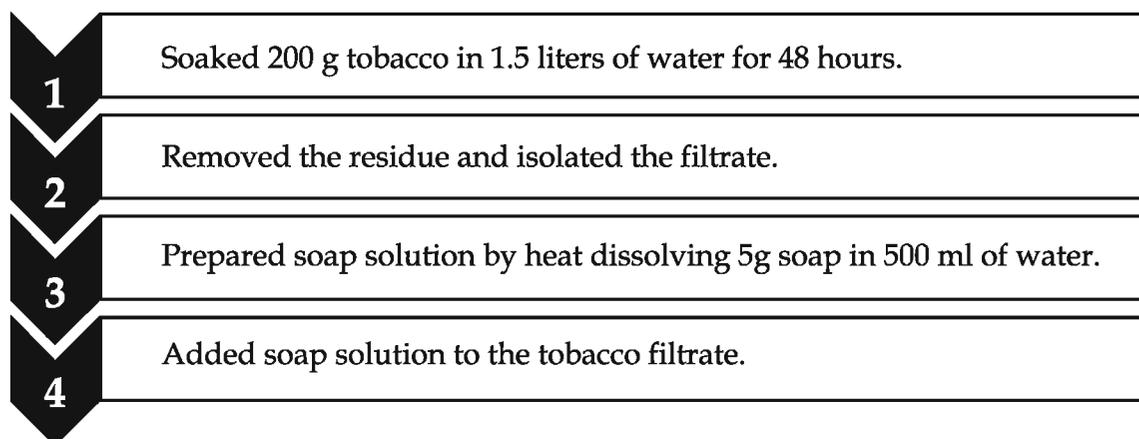
6.5.3.2.2 Planting bags

Planting bags made of Polyethylene (PE) with 35 x 40 cm dimension were purchased from M/s Bluemax Agencies Cochin.

6.5.3.2.3 Insecticides

Okra is known to be heavily affected by insects and pests such as jassid, aphids, thrips and fruit borer (Praveen *et al.*, 2007). The use of tobacco-soap emulsion has been recommended

by many authors as a safe and organic way of controlling crop pests (Chittenden, 1918 and Vijayalakshmi *et al.*, 1996). The emulsion was prepared as follows and sprayed as ten times dilutions.



6.5.3.2.4 Fertilizers

Among the three seafood based foliar formulations developed (TV100, FW100 and SH), only TV100 and FW100 were used for the study on okra, since the shrimp head (SH) foliar formulation was intended to be utilized for the color enhancement study, which would be only evident on red amaranthus. Foliar formulations were prepared as explained in Chapter 5 (5.3.3) and used for the study. The foliar fertilizers were compared with a commercially available foliar fertilizer (COM) and standard chemical soil N:P:K fertilizers such as Urea (46.1% N), Phosphate (Mussorie Rock Phosphate: 20% P₂O₅) and Potash (Muriate of Potash: 60% K₂O). Dried cow dung powder was added uniformly along with the soil in all planting bags. The chemical and organic soil fertilizer application rates were as per the recommendations of Kerala Agricultural University (KAU, 2009).

6.5.3.3 Method

6.5.3.3.1 Soil (Potting mixture) preparation

The potting mixture for the growth substratum to be filled in poly planting bags were prepared by mixing soil and dried cow dung powder in the ratio of 15:1 .

6.5.3.3.2 Planting

The okra *cv. Arka Anamika* seeds were wrapped in cloth and soaked in water for 12 hours before they were planted in the prepared planting bags. Various stages of the experiment is depicted in Plate 6.1.

6.5.3.3.3 Manures and fertilizer application

The potting mixture was kept default in all planting bags and dried cow dung powder was applied (@ 200g/plant/month) uniformly irrespective of the treatment. The seafood silage based foliar formulations such as TV100 and FW100M were applied in three concentrations such as 0.5%, 1% and 2% sprays. A wooden frame with polythene partition of the size 4 x 2 x 2 feet (Plate 6.2 (a)) was made and used to cover each plant at the time of foliar nutrient spraying in order to prevent cross contamination. The chemical NPK fertilizers were applied at the ratio

50:8:25 kg/hectare. As the experiments in this study were conducted in poly bags instead of open field, the fertilizer recommendation rates were converted to per plant rate rather than per hectare. This was done by calculating the number of plants per hectare according to the minimum area recommendation for each plant by Kerala Agricultural University (2009), which in the case of okra was 0.18 m² (i.e. 1:0.16:0.5 g NPK/plant). In one experimental unit the chemical fertilizer application rate was kept at 50% of the recommendation and was supplemented with TV100 spray @ 0.5% concentration. The control was without any foliar or chemical fertilization. The entire experiment was carried out in four replications for each treatment. Watering of the plants was done every day morning and evening throughout the study. The fertilization plan and schedule is depicted in Table 6.3.

Table 6.3: Fertilization plan and schedule of application for Okra

Fertilizer treatment	Composition	Schedule of application
F1	Chemical fertilizer (NPK100%)*	Twice/cycle
F2	Chemical fertilizer (NPK50%) + Tuna Viscera Foliar spray @ 0.5%*	Chemical fertilizer: Twice / cycle Once /week
F3	Control*	-
F4	Commercial Foliar spray @ 1%*	Once /week
F5	Tuna Viscera Foliar spray @ 0.5%*	Once /week
F6	Tuna Viscera Foliar spray @ 1%*	Once /week
F7	Tuna Viscera Foliar spray @ 2%*	Once /week
F8	Fish Waste Foliar Spray @ 0.5%*	Once /week
F9	Fish Waste Foliar Spray @ 1%*	Once /week
F10	Fish Waste Foliar Spray @ 2%*	Once /week

*Added dried cow dung powder @ 200g/plant/month

6.5.3.3.4 Harvesting

Harvesting of matured pods commenced by 25.01.2010 and ended by 17.02.2010. Pods were judged for maturity by sight and cut from the base.

6.5.3.4 Observations

6.5.3.4.1 Plant height

The plant height was measured from ground level to the tip of the main stem (Plate 6.2(b)). The average height was calculated and expressed in centimeter. The measurements were made once in two weeks. So in a single cycle there were three measurements. The first measurement was without the application of foliar fertilizers and the second and third measurements after the commencement of application of foliar fertilizers.

6.5.3.4.2 Leaf area

The leaf area measurements were made once in two weeks. Similar to plant height measurements, three readings were taken in a cycle. Leaf area was calculated as per the method suggested by Ramanujam and Indira (1978). Length (L) of the leaf was measured from base to

tip of 5 leaves/plant (Plate 6.2(c)) and the breadth (B) was measured from the middle portion of 5 leaves and the average was taken. The leaf was traced on a graph and the graph area of the leaf was calculated accordingly (Plate 6.2 (d)). The final leaf area was calculated as follows:

$$\text{Area 1} = L \times B$$

$$\text{Area 2} = \text{Graph Area (cm}^2\text{)}$$

$$\text{Constant factor} = \text{Area 2} / \text{Area 1}$$

$$\text{Leaf area} = \text{Area 1} \times \text{Constant factor}$$

6.5.3.4.3 Fruit yield

Individual fruits after harvesting were weighed and weight noted (Plate 6.2 (e)). Average fruit yield from each treatment quadruplets were expressed as grams after ending the harvest process.

6.5.3.5 Statistical analysis

The entire experiment was carried out in a Completely Randomized Design (CRD). Analysis of covariance was carried out to find the effect of different treatments on plant height and leaf area of okra. Least square means were computed for each treatment to eliminate the effect of covariate (initial plant height and leaf area) of okra and to find the significant difference between the treatment means. Analysis of variance was performed to find the effect of different treatments on the yield of okra. Treatment means were compared using Tukey's Test. All the statistical analysis were carried out using SAS 9.2. version.

6.5.4 Results and discussion

6.5.4.1 Plant height

The plant height is an important growth parameter directly proportional to the productive potential of plant in terms of fodder, grains and fruit yield and an optimum plant height is positively correlated with plant productivity (Omotoso and Shittu, 2007). The mean plant height measurements (Table 6.3) indicated that the plants fertilized with 100% chemical NPK fertilizers (F1) showed the maximum height of 101.75 centimeters followed by the F2 application (80.25 cm) consisting of 50% chemical NPK fertilizer, supplemented with 0.5% of TV100 foliar spray. The third highest plant height (71 cm) was reported for the F6 application consisting of TV100 spray at 1% concentration. The control (F3) reported the lowest plant height (37 cm). The commercial foliar spray application (F4) at 1% concentration showed a lower height value (51.75 cm) than all the seafood silage based foliar concentrations.

Table 6.4: Mean plant height of okra at the end of the cycle

Treatment	Mean Plant Height (cm)
F1	101.75
F2	80.25
F3	37.00
F4	51.75
F5	56.75
F6	71.00
F7	69.75
F8	53.25
F9	60.50
F10	65.00

Based on the statistical analysis of covariance, the initial plant height did not have significant effect on the final plant height of okra, whereas the average plant height of different applications was significantly different at 1% level of significance (Appendix 3, Table 6.1). The maximum plant height achieved by F1 was statistically in par with F2 and the same was significantly different from all other treatments.

Gomaa *et al.* (2005) has reported highly significant increases in vegetable plant height for potato in comparison with the control treatment and recommended NPK doses when organic fertilizers were applied through foliar treatment. In this experiment, the initial rate of increase in plant height in percentage was observed maximum in seafood silage based foliar formulations when compared with control, commercial foliar spray and recommended dosage of chemical NPK fertilizers. The first readings after the commencement of fertilizer applications indicated that all the seafood foliar formulations from F5 to F10 showed up to 100% increase in the rate of plant growth, while the NPK applied plants showed only around 90% increase. But during the subsequent measurements it was observed that the chemical NPK fertilizer applied plants maintained the growth rate while the organic seafood foliar sprayed plants showed a reduced trend. The experiment group applied with 50% of recommended NPK (F2) and supplemented with 0.5% TV100 spray showed a constant rate of increase throughout the study period within the range of 75 and 95%. From this it is clear that the increase in the plant height was only during the initial phase for the organic seafood foliar applied plants while the chemical NPK applied plants gained maximum growth during later stages. Table 6.5 depicts the

rate of increase in plant height in percentage between the measurements.

Table 6.5 Rate of increase in plant height of okra between the measurements.

Treatments	2nd Measurement (%)	3rd measurement (%)
F1	93	142
F2	74	96
F3	59	17
F4	65	34
F5	81	38
F6	111	48
F7	100	52
F8	99	25
F9	98	46
F10	88	44

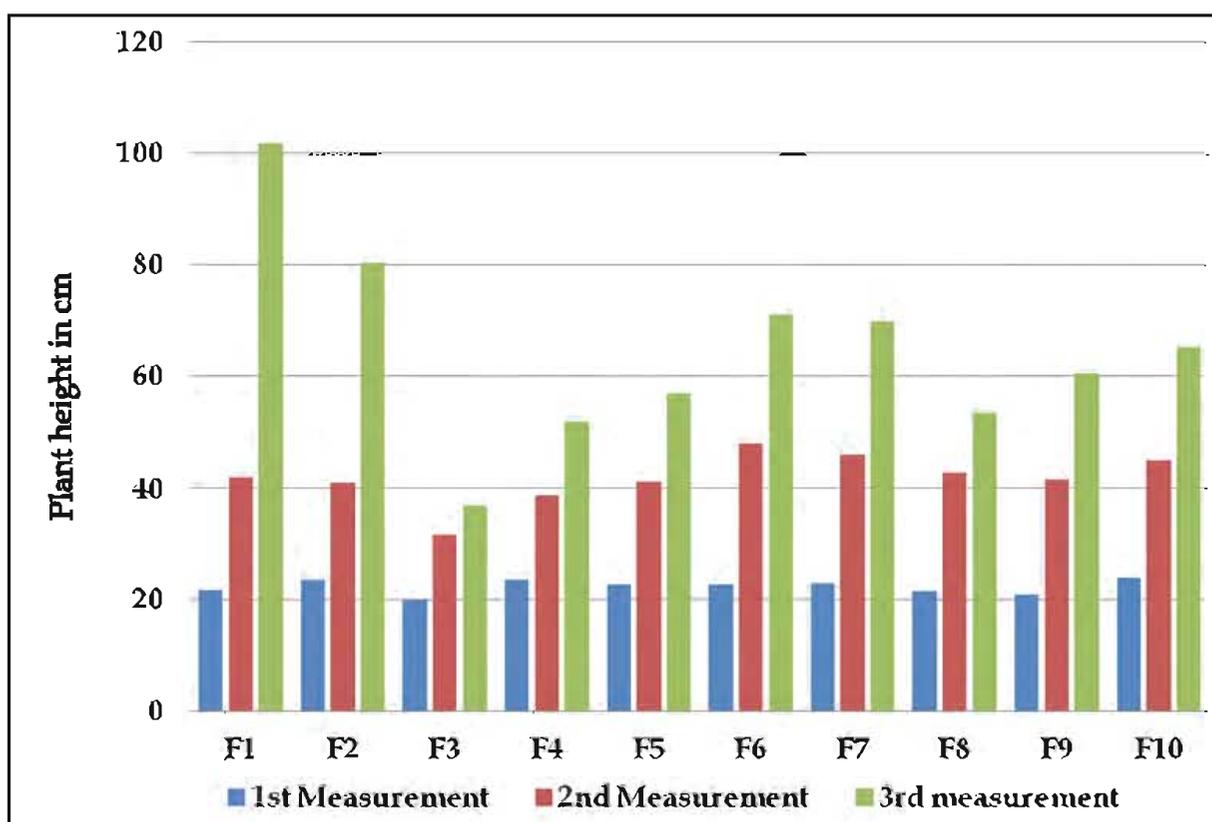


Fig. 6.2: Comparison of increase in okra plant height between the measurements (cm)

This observation agrees with the findings of Abbasi *et al.* (2010) that the chemical NPK fertilization generated a maximum height response in okra plants. The study also proves that a steady plant growth in terms of height could be achieved also at 50% of recommended NPK when supplemented with foliar fertilization. Fig. 6.2 depicts the comparison of increase in okra plant height from the first measurement to the last one between the measurements.

6.5.4.2 Leaf area

Ogoke *et al.* (2003) indicated that surface area of the leaves exposed to solar radiation is an important factor for photosynthesis, which is one of the key factors determining the plant productivity and thus it is an important component in crop growth analysis. The mean leaf area of the initial, second and third measurements (Table 6.6) indicated that the plants fertilized with 100% chemical NPK fertilizers (F1) showed the maximum area of 430 cm² followed by F2 (293 cm²) consisting of 50% chemical NPK fertilizer, supplemented with 0.5% of TV100 foliar spray. The third highest leaf area (198.5 cm²) was reported by the F9 application consisting of FW100 spray at 1% concentration. The control (F3) reported the lowest leaf area (98 cm²) while commercial foliar spray application (F4) at 1% concentration showed a value of 153 cm² which was higher than only F8 consisting of fish waste foliar spray at 0.5% concentration.

Table 6.6: Mean leaf area of Okra at the end of the cycle

Treatment	Mean Leaf Area (cm ²)
F1	430
F2	293
F3	98
F4	153
F5	163
F6	184
F7	176
F8	146
F9	198
F10	195.5

Based on the statistical analysis of covariance, the initial leaf area did not have significant effect on the final leaf area of okra, where as the average leaf area of different applications was significantly different at 1% level of significance (Appendix 3, Table 6.2). All the treatments were found to be significantly different from each other in the case of mean leaf area.

In the case of rate of increase in okra leaf area, the initial readings did not show any difference between the groups. All the experimental groups reported about 100% increase in leaf area when compared with the first measurement. All the experimental groups showed a higher increase in leaf area compared to the control (F3). Fig. 6.3 depicts the comparison of increase in okra plant leaf area from the first reading to the third one between the measurements.

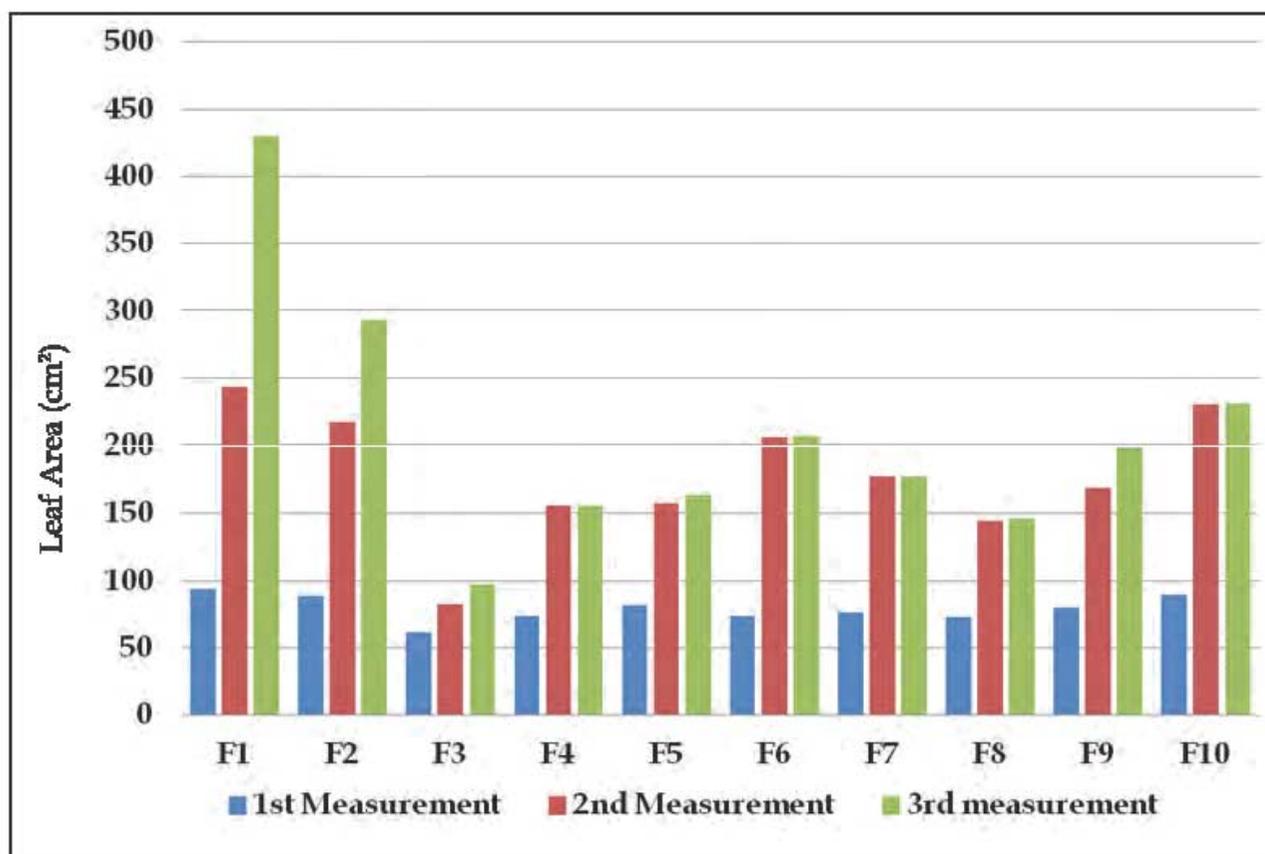


Fig. 6.3: Comparison of increase in okra plant leaf area between the measurements (cm²)

When the rate of increase between the second and third leaf area readings were analyzed, it was observed that the increase rate in F1 and F2 reduced almost half than that of the first observation, the rest of the groups from F3 to F10 indicated a very low rate. Here also like the plant height, the foliar formulations generated only an initial increase which slowed down later drastically. Table 6.7 depicts the rate of increase in plant leaf area in percentage between the treatments.

These observations were in par with the studies of Abu-Sarra *et al.* (2008) who observed that plant growth, as measured by the leaf area was considerably higher in the fertilized treatments than in the non-fertilized control and the foliar spray receiving plants showed leaf area values comparable to that of the high Nitrogen fertilizer applied units but significantly higher than those of the low Nitrogen fertilizer applied units. Khosa *et al.* (2011) also reported that macronutrients present in foliar sprays are capable of significantly increasing the leaf area in vegetables when compared to controls.

Table 6.7 Rate of increase in plant leaf area between the treatments (%).

Treatments	2 nd measurement	3 rd measurement
F1	159	76.9
F2	145	35.3
F3	34	18.8
F4	112	0.2
F5	93	3.6
F6	178	0.2
F7	132	0.3
F8	99	1.3
F9	111	17.5
F10	158	0.2

6.5.4.3 Fruit yield

The ultimate purpose of any fertilizer application is to achieve desired level or maximum productivity of agricultural crops. Babatola (2006) observed that Nitrogen as well as Phosphorus plays an important role in fruit development in okra. The mean fruit yield observations indicated that other than the plant height and leaf area observations the maximum yield of 236 g was produced by the F2 consisting of 50% chemical NPK fertilizer, supplemented with 0.5% of TV100 foliar spray. The chemical NPK fertilizer group (F1) reported the second highest yield of 232.5g (1.5% lower). All the seafood foliar concentrations reported a significantly higher yield than the control (F3) and commercial foliar spray (F4). Among these the highest yield was 178 g (140% more than F4) for F6 consisting of TV100 spray at 1% concentration. The control (F3) and commercial applications (F4) reported only 73 g and 74 g of yield respectively. The important observation of the yield study is the nominally higher okra yield generated by the F2 application unlike the pattern visible in okra plant height and okra leaf area. This observation proves that even at 50% of recommended chemical NPK dosage, it is possible to generate a similar or higher yield when complemented with foliar fertilization, which is in agreement with the observations made by Abbasi *et al.* (2010) on okra. It was noted that all the seafood silage based foliar formulations (F5 – F10) were 47 -140% better in terms of yield when compared with commercially available foliar fertilizer (F4). Even though the commercial application generated better plant height and leaf area, it could produce only identical yield obtained from the control (F3). Alkaff and Hassan (2003) also reported improved yield traits of okra in relation to foliar fertilization.

The statistical analysis of variance also indicated that the treatments were significantly different at 1% level of significance in terms of yield (Appendix 3, Table 6.3). Treatment means of yield showed that treatment F2 got maximum yield (236.5 g) and it was significantly different from other treatments. The lowest yield was produced by treatments F3 and F4, and both were statistically homogenous (Table 6.8).

Table 6.8: Mean comparison of Okra yield at the end of the cycle

Treatment	Mean Yield (g)
F1	232.50 ^B
F2	236.00 ^A
F3	73.00 ^I
F4	74.00 ^I
F5	137.50 ^F
F6	178.00 ^C
F7	127.50 ^G
F8	109.00 ^H
F9	153.50 ^E
F10	162.00 ^D

Treatment means with common letters are homogenous.

6.6 Section B

Study on the effect of seafood silage based foliar formulations on biometrical and yield characteristics of Red Amaranth (*Amaranthus tricolor* L.)

6.6.1 Introduction

The genus *Amaranthus* (Family Amaranthaceae) and its members are widely distributed throughout the world's tropical, subtropical, and temperate regions. The genus contains about 60 species and their growth habits vary from prostrate to erect, and branched to unbranched; leaf and stem colors range from red to green, with a multitude of intermediates; and seed colors range from black to white. Varieties of this species are native to a large part of India and to the islands of the Pacific and China. The plants are succulent, low growing, and compact, with growth habits much like spinach. It can be produced as a hot-season leafy vegetable in arid regions when few other leafy greens are available.

In India a number of domesticated forms are available, especially in Andhra Pradesh, Karnataka, Tamil Nadu, and Kerala. Some ornamentals with very beautiful foliage also belong

to this species. In Kerala, it is a popular leafy vegetable and is cultivated throughout the year. It is considered to be a cheap leafy vegetable in the market and it could rightly be described as the 'poor man's spinach, which is a short duration crop that fits well with the crop rotations of Kerala (Chitra and Anith, 2009). It is also found to be rich in ascorbic acid and beta-carotene and is a comparatively cheap source of nutrition for fighting malnutrition and blindness (Yadav and Sehgal, 1999). The leaves are also rich in protein, vitamins and minerals (Allemann *et al.*, 1996).

6.6.2 Amaranth fertilizers

Preetha *et al.* (2005) highlighted in their studies, the synergistic effects of combined application of organic fertilizer and chemical fertilizers in amaranth production. Amaranthus is considered as a crop which requires a high level of fertilization, and a number of field trials have shown that amaranthus has a good response to NPK fertilizers with high potassium content (Grubben, 1976). Manoharan *et al.* (2011) observed that foliar application of some bio-fertilizers improved plant productivity in amaranthus. Makus (1986, 1992) observed that sufficient soil P and supplemental N can increase *Amaranthus tricolor* L. leaf blade chlorophyll, protein and plant biomass. Foliar spray of earthworm urine was found to improve *Amaranthus* growth performance and yield (Olugbemiga *et al.*, 2011). Venkatalakshmi *et al.* (2009) observed that foliar spray of panchakavya, an organic nutrient preparation, resulted in significantly higher growth and green leaf yield. Gopalakrishnan (2007) also stated that in leaf vegetables like Amaranth, foliar application of urea is an established practice for high yield.

6.6.3 Materials & methods

Field experiment was conducted to study the effect of seafood silage based liquid foliar formulations on the Red Amaranth *cv. Arun*. The details of the materials used and methods adopted during the course of investigation are given below.

6.6.3.1 General Description

6.6.3.1.1 Location of experimental site

The field experiment was conducted in the School of Industrial Fisheries, lake side campus, Cochin University of Science & Technology, Cochin, Kerala during April 2010 – June 2010.

6.6.3.1.2 Climatic and weather conditions

The monthly meteorological data during April 2010 – June 2010 was recorded from the Meteorological Observatory of Indian Meteorological Department, Trivandrum Regional Branch and is given in Table 6.9. The mean temperature ranged from 23.9°C (June) to 32.6°C (April), while the mean monthly relative humidity ranged from 77.1% (April) to 90.4% (June). The entire cropping period received 43.8 cm rainfall from April to June.

6.6.3.1.3 Soil characteristics

The soil used for the experiment was medium deep brown. A composite soil sample was taken from the experimental area and was analyzed for physical and chemical properties and the values obtained are furnished in Table 6.10.

Table 6.9: Monthly meteorological data for April 2010 – June 2010

	Months	Temperature (°C)		Relative Humidity (%)	Mean Rainfall (mm)
		Mean minimum	Mean maximum		
1.	April 2010	24.3	32.6	77.1	4.9
2.	May 2010	25.7	33.2	82.6	10.4
3.	June 2010	23.9	31.3	90.4	28.5

Table 6.10: Physico- chemical properties of soil

Component	Value	Reference
A. Physical properties		
Sand (%)	39.5± 1.6	Pipette method (Day, 1965)
Clay (%)	8.2± 0.56	Pipette method (Day, 1965)
Silt (%)	52.3±0.68	Pipette method (Day, 1965)
B. Chemical properties		
Total N (kg/ha)	283± 1.2	Kjeldahl method (Subbaiah and Asija, 1956)
P (mg/kg soil)	75.2± 1.6	Olsen's method (Jackson, 1967)
K (mg/kg soil)	79± 0.76	Flame photometric method (Jackson, 1967)
Organic carbon %	2.1± 0.54	Wet oxidation method (Jackson, 1967)
pH	6.9± 0.65	pH meter (Piper, 1966)

6.6.3.2 Materials used

6.6.3.2.1 Seed

The Amaranth cv. Arun seeds were purchased from the Central Nursery of Kerala Agricultural University, Thrissur. This variety was introduced in 1992 by KAU through mass selection of Palapoor local, having maroon red leaves with its high yielding and photo insensitive nature. It is suitable for multi cut and is adapted to hot humid climatic conditions. It could be cultivated around the year.

6.6.3.2.2 Polyhouse

A poly house of 10x5x7m was constructed using wooden poles and polyethylene (PE) sheets to provide controlled condition for foliar spray experiment (Plate 6.3 (a)).

6.6.3.2.3 Planting bags

Polyethylene bags of 35 x 40 cm dimension were purchased from M/s Bluemax Agencies, Cochin for planting (Plate 6.3 (b)).

6.6.3.2.4 Insecticides

As a precautionary measure tobacco-soap emulsion was prepared as explained in section 6.5.3.2.3 and was applied at regular intervals (once in a week).

6.6.3.2.5 Fertilizers

Foliar formulations from tuna viscera silage (TV100) and deep-sea shrimp head silage (SH) were used in this study. The foliar preparation from fish waste (FW100) was excluded in

the study on amaranthus as from the study on okra it was clear that the maximum growth effect was resulted from TV100. The foliar formulations were prepared as explained in Chapter 5 (5.3.2) and used for the study. The shrimp head silage was prepared at 100% (SH100) and at different combinations as explained in Chapter 5 (5.3.2), with TV100, and subjected to different treatments to analyze the hypothesis of possible color enhancement of amaranth leaves due to pigments in SH silage. The foliar fertilizers were compared with standard chemical N: P: K and commercial foliar spray fertilizers.

6.6.3.3 Method

6.6.3.3.1 Soil (Potting mixture) preparation

The potting mixture were prepared by mixing soil and dried cow dung powder to the ratio of 15:1 and filled in poly planting bags.

6.6.3.3.2 Planting

The Amaranth *cv. Arun* seeds were sowed in raised seed beds and spread a thin layer of fine sand on top. The saplings of 20-30 days old having 4-5 leaves were later transplanted to poly bags.

6.6.3.3.3 Manures and fertilizer application

The potting mixture was kept default in all planting bags and dried cow dung powder was applied (@ 200g/plant/month) uniformly irrespective of the treatment. From the results of the okra, it was evident that maximum yield among the foliar formulations was obtained from 1% concentration of TV100. Hence the seafood silage based foliar formulations were applied at 1% concentrations uniformly in amaranth study also. The wooden frame with polythene partition to prevent cross contamination during spraying was used as in okra experiment.

The chemical NPK fertilizers were applied at the ratio 100:50:50 kg/hectare. Like in okra, the fertilizer recommendation rates were converted to per plant rate rather than per hectare, by calculating the number of plants per hectare. Thus the minimum area for each plant as recommended by Kerala Agricultural University (2009) was 0.15 m² (i.e. 14.8:7.4:7.4 g NPK/plant) in the case of Amaranthus.. In two experimental units the chemical fertilizer application rate was kept at 50% and 25% of the recommended dose and was supplemented with TV100 spray @ 1% concentration.

In this experiment, the shrimp head (SH) and Tuna viscera (TV100) combinations in different ratio were made and subjected to treatments by controlling oxidation, exposure to light and temperature (OLT treatment) as explained in Chapter 5 (5.3.2) with the main objective of preserving the natural pigments in SH silage. Owing to the presence of comparatively low concentration of digestive enzymes in SH head waste; it was mixed with TV100 in different concentrations for ensuring accelerated ensilation. The plants were sprayed with treated and untreated sprays @ 1% concentration in order to compare the results. The control (F4) was without any foliar or chemical fertilization. The entire experiment was carried out in quadruplets per treatment. The chemical and organic soil fertilizer application rates were as per the recommendations of Kerala Agricultural University (KAU, 2009). Watering of the plants

was done every day morning and evening throughout the period. The fertilization plan and schedule is depicted in Table 6.11.

Table 6.11: Fertilization plan and schedule for Amaranthus

Fertilizer treatment	Composition	Schedule of application
F1	Chemical fertilizer (NPK100%)*	Twice/cycle
F2	Chemical fertilizer (NPK50%)*	Twice/cycle
F3	Chemical fertilizer (NPK25%)*	Twice/cycle
F4	Control*	
F5	Commercial Foliar spray @ 1%*	Once/week
F6	Chemical fertilizer (NPK50%) + Tuna Viscera Foliar spray @ 1%*	Chemical fertilizer: Twice / cycle
		Foliar spray: Once/week
F7	Chemical fertilizer (NPK25%) + Tuna Viscera Foliar spray @ 1%*	Chemical fertilizer: Twice / cycle
		Foliar spray: Twice/week
F8	Shrimp head Foliar spray treated @ 1%*	Once / week
F9	Shrimp head Foliar spray non-treated @ 1%*	Once / week
F10	Shrimp head and Tuna Viscera treated (50/50) @ 1%*	Once / week
F11	Shrimp head and Tuna Viscera non-treated (50/50) @ 1%*	Once / week
F12	Shrimp head and Tuna Viscera treated (25/75) @ 1%*	Once / week
F13	Shrimp head and Tuna Viscera non-treated (25/75) @ 1%*	Once / week
F14	Shrimp head and Tuna Viscera treated (75/25) @ 1%*	Twice/week
F15	Shrimp head and Tuna Viscera non-treated (75/25) @ 1%*	Twice/week

*Added dried cow dung powder @ 200g/plant/month

6.6.3.3.4 Harvesting

The matured amaranth foliage was harvested by cutting the entire shoot at the height 10 cm from the bottom (Plate 6.3 (c)).

6.6.3.4 Observations

6.6.3.4.1 Plant height

The plant height was measured similar to that of okra as explained in 6.5.3.4.1.

6.6.3.4.2 Leaf area

The leaf area was measured similar to that of okra as explained in 6.5.3.4.2.

6.6.3.4.3 Foliage yield

Individual plants after harvesting were weighed and weight noted. Average plant foliage yield of each treatment were expressed in grams.

6.6.3.4.4 Leaf Color

Leaf color measurements were done using a Hunter lab Colorimeter Model No D/8-S (Miniscan XE Plus) with geometry of diffuse /8° (sphere 8 mm view) and an illuminant of D65/10 degree. Color of the freshly cut amaranth leaf was measured and expressed as the mean of three measurements. In the Hunter scale, L* measures lightness and varies from 100 for perfect white to zero for black, approximately as the eye would evaluate it. The chromacity dimensions (a* and b*) give understandable designations of color as follows: a* measures redness when positive, gray when zero, and greenness when negative. b* measures yellowness when positive, gray when zero, and blueness when negative.

6.6.3.5 Statistical analysis

The entire experiment was carried out in a Completely Randomized Design (CRD). Analysis of covariance was carried out to find the effect of different treatments on plant height and leaf area of *Amaranthus*. Least square means were computed for each treatment to eliminate the effect of covariate (initial plant height and leaf area). All possible pair wise comparison of least square means were also carried out to find the significant difference between the treatment means. Analysis of variance was performed to find the effect of different treatments on the yield and leaf color of *Amaranthus*. Treatment means were compared using Tukey's Test. All the statistical analysis was carried out using SAS 9.2.

6.6.4 Results and discussion

6.6.4.1 Plant height

The plant height measurements (Table 6.12) indicate that the plants fertilized with 100% chemical NPK fertilizers (F1) showed the maximum height of 53.5 centimeters followed

Table 6.12: Mean plant height of *Amaranthus* subjected to various treatments

Treatment	Mean Plant Height (cm)
F1	53.5
F2	48.1
F3	43
F4	35
F5	48
F6	45
F7	47.2
F8	48.75
F9	45
F10	50.75
F11	49.5
F12	50.25
F13	47.75
F14	43.5
F15	46.5

by the F10 application (50.75 cm) consisting of Shrimp head and Tuna Viscera treated (50/50) sprayed at 1% concentration. The third highest plant height (50.25 cm) was obtained for the F12 treatment consisting of Shrimp head and Tuna Viscera treated (25/75) sprayed at 1% concentration. The control (F4) produced the lowest plant height (35 cm) while commercial foliar spray application (F5) at 1% concentration produced a height of 48 cm.

Based on the statistical analysis of covariance, the initial plant height did not have significant effect on the final plant height of amaranthus, whereas the average plant height of different applications was significantly different at 1% level (Appendix 3, Table 6.4).

The results indicated that unlike the okra, the amaranthus plants did not show any noticeable difference in the rate of increase in the plant height. Almost all the experimental groups showed a uniform rate of increase in height (21 to 47%). The rate of increase doubled during the 3rd measurement (55 to 78%), which was also uniform in all the groups. This indicates that any form of fertilization has a positive response on amaranth plant height. Table 6.13 depicts the rate of increase in plant height in percentage between the treatments.

In this experiment, it was also noted that among the experimental treatments, only F1, F10 and F12 reported a height above 50 cm, whereas the rest of the treatments including the commercial foliar spray (except the control), produced plant height between 40 and 50 cm. Fig. 6.4 depicts the comparison of increase in plant height from the first measurement to the last one between the treatments.

Table 6.13 Rate of increase in plant height between the treatments.

Treatments	2 nd Measurement (%)	3 rd measurement (%)
F1	21	55
F2	47	65
F3	33	78
F4	65	74
F5	26	65
F6	30	72
F7	27	67
F8	25	63
F9	30	72
F10	23	59
F11	24	62
F12	24	60
F13	26	66
F14	32	77
F15	28	68

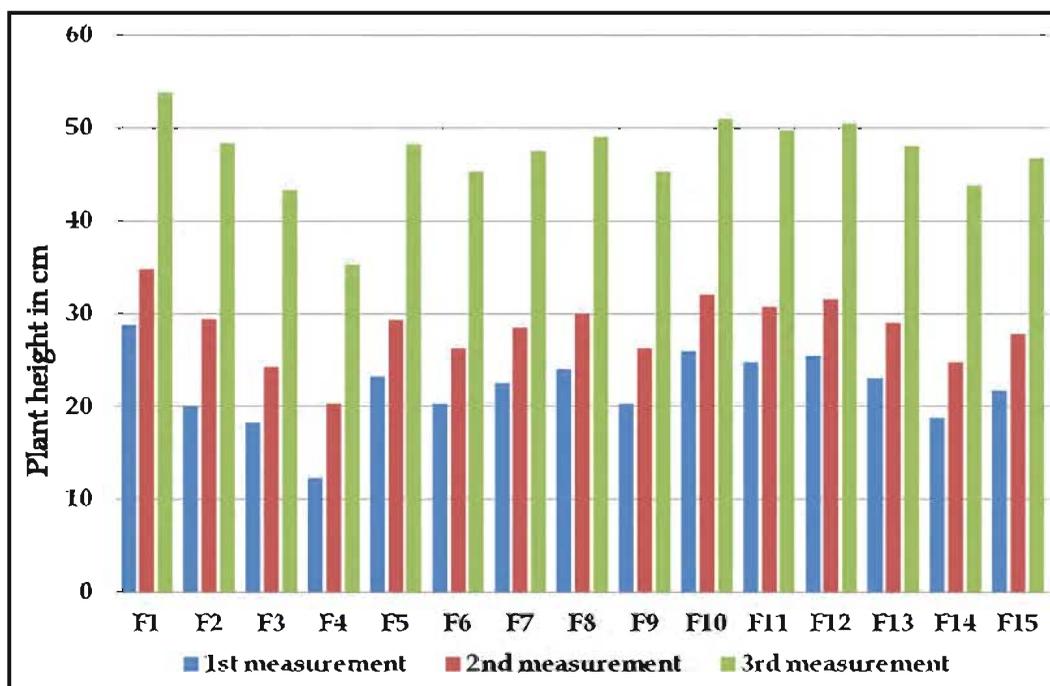


Fig. 6.4 Comparison of increase in amaranthus plant height between treatments at various stages of measurements

Seran and Brintha (2011) have reported significant effect of NPK fertilizers on plant height of *Amaranthus tricolor*. Increasing vegetative growth due to proper NPK fertilization levels has been also reported by Chen *et al.* (1996) on eggplant and Gupta and Sengar (2000) Sun *et al.* (2001) and Kotepong *et al.* (2003) on tomato plant.

6.6.4.2 Leaf area

The leaf area measurements (Table 6.14) indicated that maximum leaf area was 185.5 cm²

Table 6.14: Mean comparison of leaf area of *Amaranthus* under various treatments

Treatment	Mean leaf area (cm ²)
F1	165.2
F2	155.5
F3	150.5
F4	92
F5	135
F6	130
F7	185.5
F8	128.5
F9	120
F10	125.5
F11	173.5
F12	108
F13	155
F14	146
F15	175

obtained for F7 treatment consisting of chemical fertilizer (NPK 25%) + Tuna Viscera Foliar spray @ 1% concentration. The second and third highest leaf area was obtained for the F 15 (175 cm²) and F 11 (173.5 cm²) treatments consisting of Shrimp head and Tuna Viscera non-treated (75/25) @ 1% concentration and Shrimp head and Tuna Viscera non-treated (50/50) @ 1% concentration respectively. The control (F4) produced the lowest leaf area (92 cm²) while commercial foliar spray application (F5) at 1% concentration showed a value of 135 cm².

Based on the statistical analysis of covariance, the initial leaf area did not have significant effect on the final leaf area of amaranthus, whereas the average leaf area of different applications (treatment) was significantly different at 1% level of significance (Appendix 3, Table 6.5). All the treatments were found significantly different from each other in the case of mean leaf area.

In the case of rate of increase in amaranthus leaf area, the initial, secondary and tertiary reading showed maximum increase during initial period among the seafood foliar spray formulations, which later became more uniform and reduced, when compared with chemical NPK applications. When compared to control (F4), the seafood foliar formulations showed around 80% increase in leaf area and around 30% increase in leaf area when compared to commercial formulation (F5). Table 6.15 depicts the rate of increase in plant leaf area between the treatments at first, second and third measurements.

Table 6.15 Rate of increase in plant leaf area between the treatments.

Treatments	2 nd measurement (%)	3 rd measurement (%)
F1	12	7
F2	13	8
F3	14	8
F4	128	14
F5	248	14
F6	17	5
F7	16	28
F8	133	9
F9	112	10
F10	124	10
F11	176	7
F12	163	11
F13	244	8
F14	223	8
F15	280	7

The study of Olugbemiga *et al.* (2011) indicated that foliar application of organic extracts resulted in substantial increase in leaf area of amaranthus. Fig. 6.5 depicts the comparison of increase in amaranthus plant leaf area between measurements at various stages of growth.

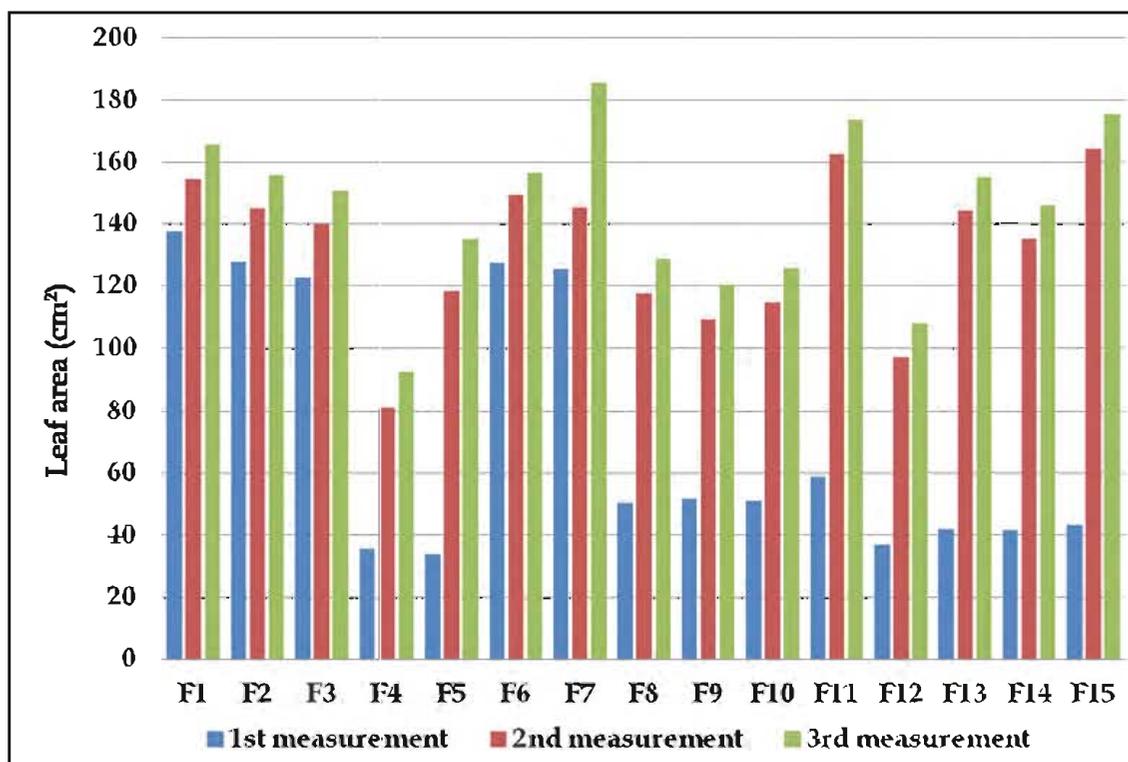


Fig. 6.5 Comparison of amaranthus plant leaf area between the treatments at various stages of growth.

6.6.4.3 Plant yield

As the consumable part of the amaranthus is the entire plant, the whole plant weight was recorded. The plant yield observations indicated that the maximum yield of 261.45 g was produced by the F7 treatment consisting of chemical fertilizer (NPK 25%) + Tuna Viscera Foliar spray @ 1% concentration. The chemical NPK fertilizer treatment (F1) produced the second highest yield of 242.85 g. All the seafood foliar concentrations reported a significantly higher yield than the control among which the highest was the F14 (132.84 g) treatment consisting of Shrimp head and Tuna Viscera treated (75/25) @ 1% concentration. The results of analysis of variance indicated that the average yield of different treatments were significantly different at 1% level of significance (Appendix 3, Table 6.6). Treatment means of yield (Table 6.16) showed that treatment F7 got maximum yield (261.45g) and it was significantly different from other treatments. In the case of other treatments, treatment F1 was homogenous with F6, treatment F10 was homogenous with F11 and F8 was homogenous with F9.

When only chemical NPK fertilizers were added at 25% of recommended dosage (F3), the plant group produced only 205.25 g of yield, which was 18% less than the 100% NPK recommendation (F1). But when the F3 was supplemented with Tuna Viscera Foliar spray @ 1% concentration, the group (F7) produced a plant yield which was 8 % more than the 100% NPK recommendation. Positive effect of organic fertilizers on vegetative growth has been observed by Raghava and Murty (1998). Preetha *et al.* (2005) has also observed that combinations of organic and NPK fertilizations produced better yield in amaranthus than single fertilizer application.

Table 6.16: Comparison of Amaranthus Yield at various treatments

Treatment	Mean Yield (g)
F1	242.85 ^B
F2	220.75 ^C
F3	205.25 ^D
F4	87.25 ^L
F5	111.68 ^I
F6	240.25 ^B
F7	261.45 ^A
F8	99.02 ^L
F9	101.25 ^K
F10	118.55 ^G
F11	117.6 ^G
F12	114.67 ^H
F13	104.39 ^J
F14	132.84 ^E
F15	129.25 ^F

Treatment means with common letters are homogenous.

6.6.4.4 Leaf color

The leaf color analysis indicated that, the a^* factor indicating the red color intensity was found maximum (8.6) in plants sprayed with Shrimp head (SH) foliar spray, treated (OLT) at 1% concentration. The second highest a^* value was produced by F11 (8.4) consisting of Shrimp head and Tuna Viscera non-treated (50/50) and sprayed at 1% concentration. Subsequently the third and fourth values of a^* were shown by F9 (7.3) and F10 (5.3) comprising of SH Foliar spray non-treated and SH and Tuna Viscera treated (50/50) sprayed at 1% concentration respectively. The b^* factor indicating the yellowness were found to be maximum in F1 and L^* values indicating the lightness was found to be maximum in F5 consisting of commercial foliar spray sprayed at 1% concentration. From the observations, the hypothesis that shrimp pigment preserved from being oxidized, had effect on amaranth leaf color enhancement is rejected. Increased redness was observed in plants applied with seafood foliar sprays, irrespective of the treatment. However maximum redness values (a^*) were exhibited by the plants sprayed with foliar sprays with SH component with or without treatment. Fig. 6.6 shows the ratio of L^* , a^* and b^* values between treatments. Plate 6.4 shows the variations in leaf colour of amaranthus sprayed with different foliar applications.

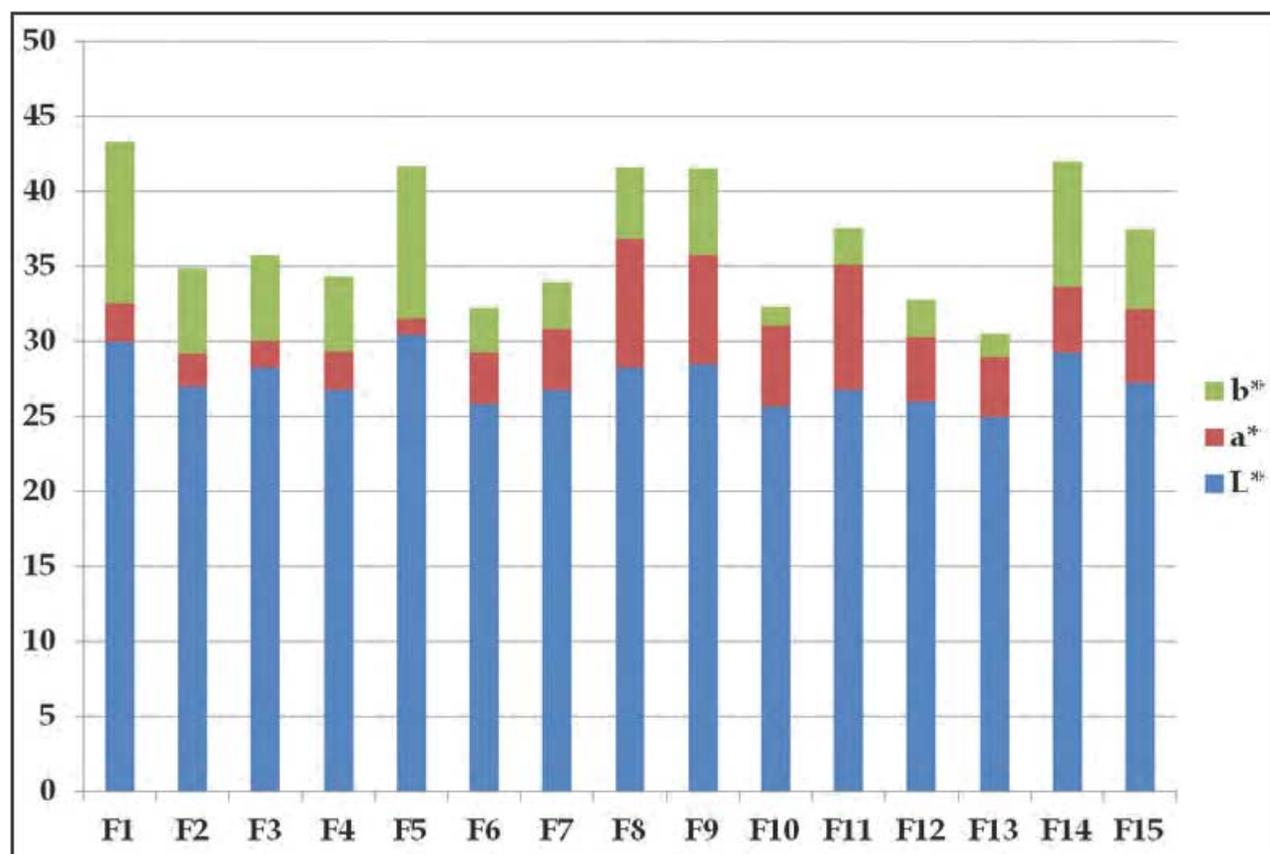


Fig. 6.6: Ratio of L*, a* and b* values between treatments

The statistical analysis of variance also indicated that the treatments were significantly different at 1% level of significance on different L*a*b* values (Appendix 3, Table 6.7, 6.8 and 6.9). Treatment mean of L* values shows that the maximum value attained by F5 application was significantly different from F10 and F13 application, while the rest were homogenous. Treatment mean of a* values shows that the maximum values were produced by F8 and F11 applications and both were homogeneous with F9, but the two were significantly different from the rest of the applications/treatments. Treatment mean of b* values shows that the maximum values attained by F1 and F5 applications and both were homogeneous with F14 and significantly different from the rest of the applications. Table 6.17 depicts the Tukey's Studentized Range (HSD) Test for color (L*a*b* values).

The red color in amaranthus leaf is attributed to the presence of Amaranthine, a purple pigment found in leaves of Amaranthus species which belongs to betacyanins, a group of water-soluble nitrogen-containing pigments (Azeredo, 2009). Betacyanin is a phytonutrient and an antioxidant and is responsible for the distinguishing deep red color of red amaranth (Khandaker *et al.*, 2009). Khandaker *et al.* (2011) has observed that foliar application with organic acids such as salicylic acid enhanced biosynthesis of photosynthetic pigments chlorophyll and bioactive compounds betacyanins, total polyphenol etc. with increased number of leaf per plant, leaf size, and fresh and dry matter yield in red amaranth. This could also be the reason for the increase in redness in the case of seafood foliar spray applied leaves, as the formulations

Table 6.17: Tukey's Studentized Range (HSD) Test for color.

Treatment	Mean L*	Mean a*	Mean b*
F1	29.983 ^{AB}	2.5667 ^{DEFG}	10.7867 ^A
F2	27.03 ^{ABC}	2.1433 ^{EFG}	5.69 ^{BC}
F3	28.3 ^{ABC}	1.73 ^{FG}	5.76 ^{BC}
F4	26.777 ^{ABC}	2.6 ^{DEFG}	5.0067 ^{BCD}
F5	30.443 ^A	1.1 ^G	10.19 ^A
F6	25.81 ^{ABC}	3.4967 ^{CDEF}	2.9433 ^{CDE}
F7	26.807 ^{ABC}	4.0633 ^{CDE}	3.11 ^{CDE}
F8	28.28 ^{ABC}	8.5967 ^A	4.7767 ^{BCDE}
F9	28.463 ^{ABC}	7.2967 ^{AB}	5.7567 ^{BC}
F10	25.673 ^{BC}	5.39 ^{BC}	1.27 ^E
F11	26.747 ^{ABC}	8.4133 ^A	2.3667 ^{CDE}
F12	26.02 ^{ABC}	4.2367 ^{CD}	2.57 ^{CDE}
F13	24.99 ^C	4 ^{CDE}	1.5267 ^{DE}
F14	29.28 ^{ABC}	4.36 ^{CD}	8.36 ^{AB}
F15	27.227 ^{ABC}	4.91 ^C	5.3367 ^{BC}

Treatment means with common letters are homogenous.

contains formic acid which is also an organic acid. This could be attributed to the maximum redness shown by the plants applied with SH incorporated foliar sprays since the amount of formic acid added to prepare SH based silages was higher (7%) than that of TV (3.5%). Another important factor responsible for enhanced red pigment concentration in the leaves sprayed with seafood foliar fertilizers is the efficiency of nutrient absorption in foliar fertilization compared to soil fertilization and the effective nitrogen balance present in seafood foliar sprays. Studies conducted by Ali and Shinya (2011) has indicated that nitrogen fertilization significantly influenced the leaf color parameters, like redness (a*) or greenness and the trend was similar to the trend in yield production and pigments accumulation in red amaranth leaf.

6.6.4.5 Input cost economics of seafood foliar fertilization

The cost of various input factors in agriculture like fertilizers, pesticides, labor, energy, land and seed is increasing over the period in India (Wilmot, 2009). Comparing the input cost of seafood foliar preparations with that of solid chemical NPK fertilizers, it is appropriate considering the difference in physical state of both and mode of application. But the effects of seafood based foliar preparations are compared in this study with solid NPK fertilizers and commercial organic foliar formulations to emphasize the importance of the same.

When the unit production cost of seafood foliar fertilizer was calculated (Table 6.18), it was found that the possible selling price of seafood foliar formulations is around ₹30/liter,

Table 6.18: Production cost of foliar fertilizer from fish waste for a WSHG production unit

Land & Building	
Area (sq. ft.)	1000
Rental/month (₹)	15000
Equipments	
Plastic Utensils (₹)	50000
filter press (₹)	120000
Weighing balance (₹)	10000
Total fixed cost (₹)	195000
Wages/month	
Unskilled (2) (₹)	18000
Working capital	
Raw material (30t/month) (₹)	240000
Formic acid (1050L/month) (₹)	52500
Water cost/month (₹)	1000
Electricity 200 units/month (₹)	5000
Packaging material /month (₹)	72000
Production cost/month (₹)	370500
Depreciation @10% (₹)	1500
Interest on capital @ 10% (₹)	4670
Total cost of production (₹)	376670
Unit cost of production (₹)	15.69
Expected selling price (₹)	30

when it is produced on a small scale by a woman self-help group (WSHG) initiative, whereas the commercially available organic foliar sprays cost around ₹ 300-500/liter. In the case of farming of vegetables like amaranthus, it has been suggested that the area required by a single plant in the field is about 0.18 m² (Mortley *et al.*, 1992). When converting to a hectare area of

10000 m², it could be assessed that a hectare could contain around 55555 plants. During the foliar spray application studies, it was found that around 15 ml of spray is required to uniformly wet the entire leaf surface area of a plant. As the recommended dosage of seafood foliar spray is 1% for each spraying, a single plant consumes around 0.1 ml of seafood foliar formulation. Hence an entire hectare of plant on a single application consumes around 8 liters of foliar formulation at 1% dilution. When calculated in a cycle of application of okra and amaranthus it requires around 100 liters of crude seafood foliar formulation costing around ₹3000. In the case of chemical NPK fertilizers, Urea, Superphosphate and Potash costs only ₹6, 10 and 6.5 per Kg respectively on a subsidized rate of 67% currently. The cost of chemical fertilizer input for okra and amaranth is only around ₹1500 per cycle.

Considering the productivity factor in tonnes per hectare of okra, (Fig. 6.7) the NPK applied plants produced a yield of around 12.9 tonnes per hectare while the plants subjected to 50% chemical NPK and 1% seafood foliar treatment produced a 1.5% higher productivity (13.1 t/ha). Compared to the commercial foliar spray, the seafood foliar application produced more than 100% increase in productivity and it was far behind the productivity values obtained for chemical NPK fertilizer (9.8 t/ha).

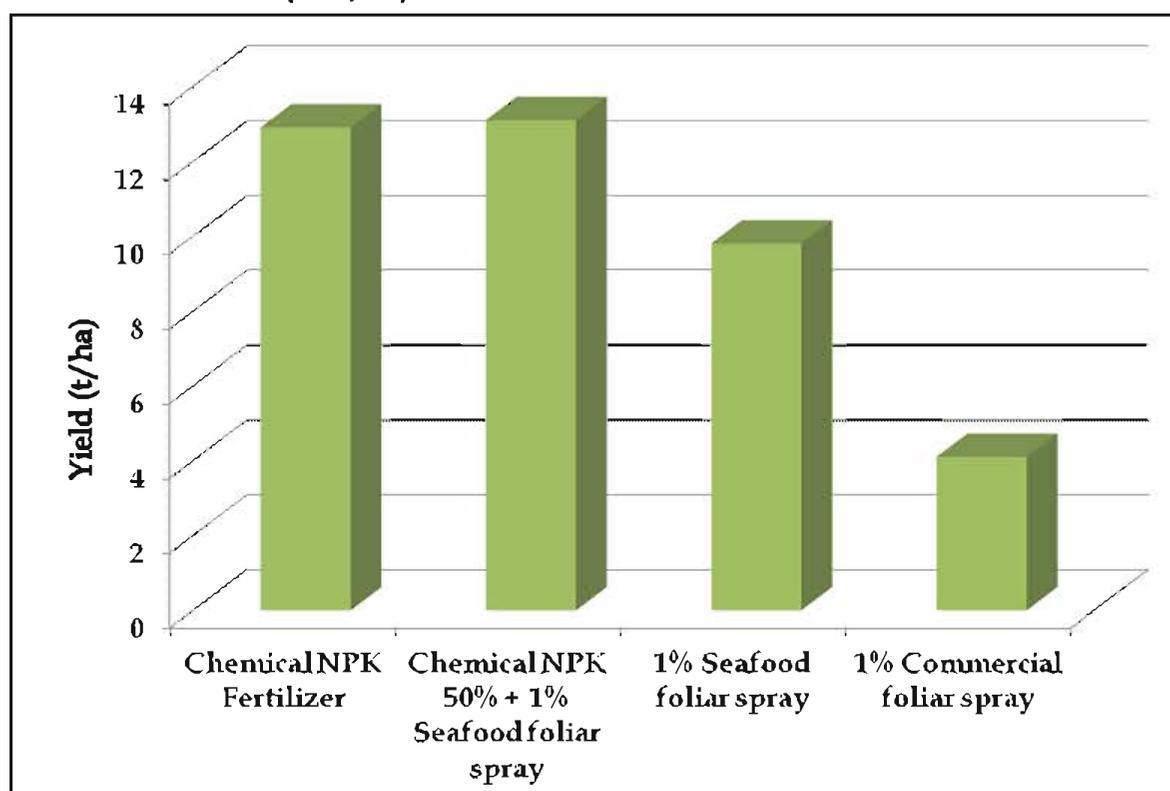


Fig. 6.7: Comparison of okra productivity between selected treatments

In amaranthus, (Fig. 6.8) the plants treated with 25% of chemical fertilizer and supplemented with 1% seafood foliar fertilization reported around 7.5% more productivity (14.5 t/ha) than the 100% chemical fertilization (13.49 t/ha). When compared with commercial foliar fertilizer, the seafood foliar spray applied plants produced around 17.7 % increase in productivity.

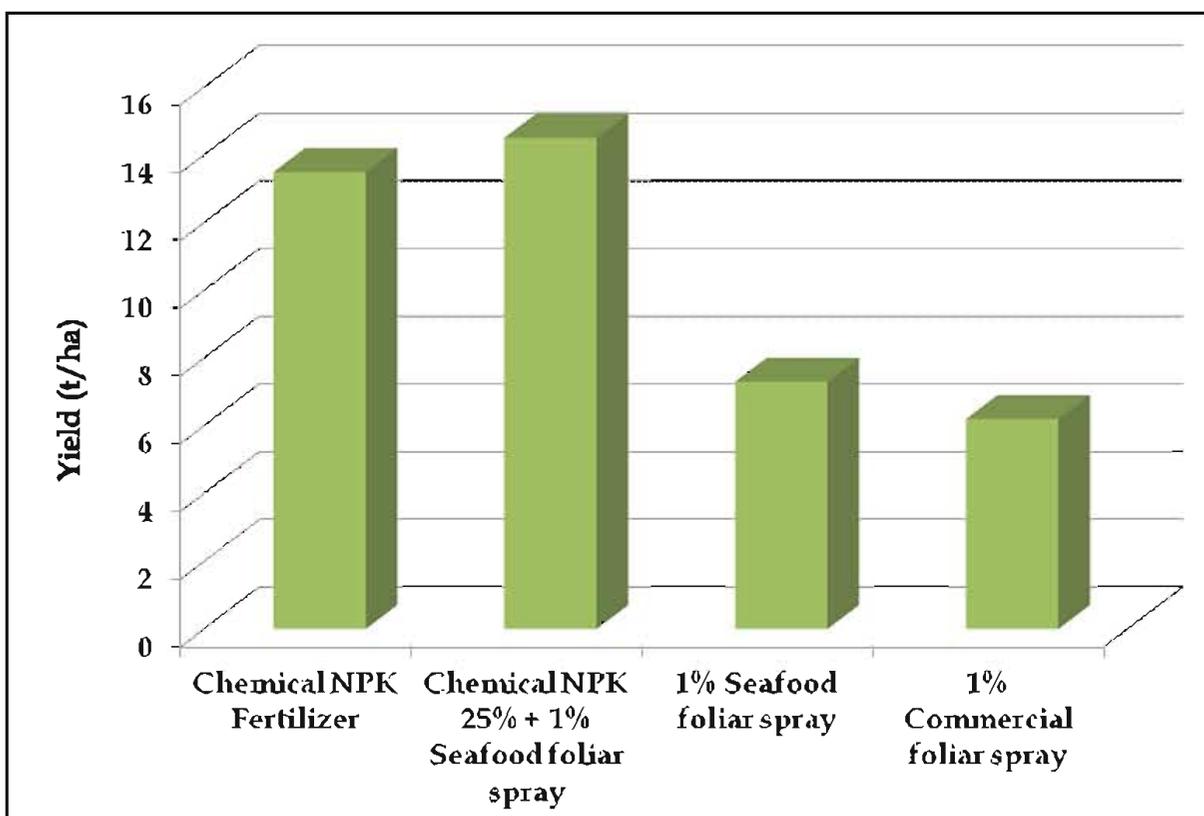


Fig. 6.8: Comparison of amaranthus productivity between selected treatments

It is clear from the results that in combination with reduced (50-75%) chemical fertilization, the foliar fertilization from seafood waste was successful in generating an increased productivity, compared with the components applied alone.

With the increased labor cost of application, the entire idea of organic farming seems to be a costly affair. Most studies also point to the fact that organic agriculture requires significantly greater labor inputs than conventional farming (World Bank, 2004). The studies has also indicated that the productivity of organic farming may be lesser than that of conventional or inorganic farming in the initial years while the yields increases progressively under organic farming on a long run. The yields of different crops in low cost sustainable system or organic farming are comparable to that of conventional system, given time for the soil conditions and the farmers to adjust and cope up (Bhattacharya and Chakraborty, 2005).

It has to be noted that the reduced cost in chemical fertilizer application is due to the heavily subsidized agricultural input policy followed by the Government. Being the most expensive aspect of India's food and agriculture policy regime, the input subsidies consume a large budget share of India. Government of India pays fertilizer producers directly, in exchange for the companies selling fertilizer, at lower price than market prices, thus resulting in subsidies of 40 to 75 percent to the farmer for fertilizer. This has distorted trade by increasing net exports of input intensive commodities, and decreasing net exports of commodities which require relatively fewer inputs (Grossman and Carlson, 2011). This pattern of subsidies has created ill effects of overutilization of inputs (chemical fertilizers and pesticides) leading to soil degradation,

soil nutrient imbalance, environmental harm and groundwater depletion, all of which have caused decreased effectiveness of inputs (Planning Commission, 2006). Considering these factors, a shift to organic way of farming is inevitable in the future for sustainability, evident from the increasing market for organic products.

Against the 2-3% growth in the conventional food industry, the organic food industry has been experiencing an annual growth between 17% and 22% over the past several years (Muthukumar, 2006). Another positive factor which is going to favor organic farmers is the huge price difference (50-100%) between the organic and conventional products in the export market, even though the domestic demand is less. But the strategy should be to begin the practice of organic farming for low volume high value crops such as spices, medicinal plants, fruits and vegetables (Swaminathan, 2007) which has a huge export demand. Organic tea, spices and fruits for example is suitable to be identified as a candidate for seafood foliar formulation based fertilization, considering the input costs and the return.

The Maharashtra State Co-operative cotton Growers Marketing Federation Ltd. (MAHACOT) in their market report, 2012 states that India exported about 300 organic products fetching ₹1,960 crore in 2011 and the Agricultural and Processed Food Products Export Development Authority (APEDA) has fixed a target of touching ₹5,000 crore in organic products exports by 2015. This report continues to state that around 4.84 million hectares are under organic farming in India and only 16 per cent of organic products produced in the country are exported. The Indian organic products are primarily exported to Europe, US, Canada and Japan. In Europe, Germany and Switzerland are the main buyers. It is evident that the Indian organic sector is climbing the development ladder on a faster rate than the conventional agricultural sector. But certain steps need to be taken on a long run to ensure the growth in a sustainable manner such as equipping small farmers/NGOs and traders for successful organic farming and marketing, standardizing organic certification procedures, creating a reliable database on organic product availability, strengthening organic rules and regulations and educating domestic customers regarding the benefits of organic products (Garibay and Jyoti, 2003). The present study proves that the production and usage of foliar spray formulations from seafood waste can greatly augment the organic agriculture scenario.

6.7 Conclusion

Okra

- This study recommends that even at 50% of recommended chemical NPK dosage, it is possible to generate a similar or higher yield when complemented with foliar fertilization TV100 @ 1% concentration.
- All the seafood silage based foliar formulations were 47 -140% better in terms of yield when compared to commercially available foliar fertilizer. The commercial foliar spray, even though generated better plant height and leaf area, could produce only identical yield data with that of control.

Amaranthus

- Plants treated with 25% of the recommended chemical NPK dosage supplemented with Tuna Viscera Foliar spray @ 1% concentration, produced a plant yield which was 7.7 % more than the 100% NPK application.
- Seafood foliar spray consisting of shrimp head and Tuna viscera treated (75/25) and sprayed @ 1% concentration produced a 52% higher yield than the commercial foliar formulation. This proves that the seafood foliar spray provided a more balanced nutritional supply for the plants than other treatments.
- Leaf colour analysis indicated that the seafood foliar spray consisting of shrimp head silage and sprayed @ 1% concentration generated a 200% higher redness value (a*) than the commercial foliar formulation and chemical NPK treated plants.

Economics

- Even though higher in terms of input and labour costs when compared to the heavily subsidised chemical fertilizer cost, which also cause environmental and economical ill effects by the unregulated usage, it could be recommended that the seafood foliar fertilization, in the long run, may be adopted as a reliable organic agricultural input.
- Utilization of seafood processing waste based foliar fertilization as an organic fertilizer input could be a practical solution for addressing the waste management issues faced by the seafood industry as it reduces the waste management cost and subsequent impact of waste on the environment.





6.1(a): Planted bags arranged for the experiment



6.1(b): One week old okra seedling



6.1(c): Experimental okra plants

Plate 6.1: Various stages of planting the okra seedlings



6.2(a): Wooden frame to prevent cross contamination of foliar applications



6.2(b): Measurement of okra plant height



6.2(b): Measurement of okra plant length



6.2(d): Leaf outline taken for calculating leaf area



6.2(e): Harvested okra pods

Plate 6.2: Measurement of various parameters of okra



6.3(a): Poly house for amaranthus study



6.3(b): Experimental amaranthus plants



6.3(c): Harvested amaranthus foliage

Plate 6.3: Various stages of amaranthus growth study



6.4(a): Leaf colour of amaranthus sprayed with commercial (COM) foliar spray



6.4(b): Leaf colour of amaranthus applied with chemical fertilizer (KAU100)



6.4(c): Leaf colour of amaranthus sprayed with tuna based (TV100) foliar spray



6.4(d): Leaf colour of amaranthus sprayed with shrimp head based (SH100) foliar spray

Plate 6.4: Variations in leaf colour of amaranthus applied with different formulations

CHAPTER 7

SUMMARY & CONCLUSION

Waste management as a term has intrigued the human civilization since the beginning of production, storage and consumption of goods. Several technologies were introduced for the management of waste generated, among which the biodegradable waste or the wastes produced from the organic sources were of prime importance. As time passed, there was an increase in the percentage of organic waste production due to inefficient harvesting, pre-processing, processing, transportation and value-addition. As a result, the land and water were overwhelmed with organic matter both from industrial and domestic discharge. Thus the organic pollution from food industry and household has become one of the major environmental issues which needs immediate attention. This study discusses the impact of one of the important and critical form of organic waste, that is from seafood, which acts as a major contributor of protein diet of the world.

In **Chapter 1**, a general introduction on the issue of seafood waste management was given in light of the growing concern on the pollution potential of the seafood processing sector in India. The scope and significance of the study was discussed as it pioneers an issue based analysis of seafood waste management in Aroor Seafood Industrial Belt (ASIB) Kerala, one of the important seafood processing zones in the country along with development of a suitable by-product from seafood waste. The objectives of the study were mentioned as determination of the current status of seafood waste generation, management and utilisation in the area, assessment of the extent of impact generated by the current waste management practices over the coastal community, analysis of the nutritional quality of seafood waste, development of foliar fertilizer formulations from seafood waste and studying their effect on plant varieties such as okra (*Abelmoschus esculentus*) and red amaranth (*Amaranthus tricolor* L.).

The **Chapter 2** reviewed the pollution potential of seafood processing wastes by indicating the key environmental issues of concern regarding the waste generation from seafood processing industries. This chapter also presents an extensive review of literature on the status of seafood waste management methods currently adopted worldwide, for both solid and liquid waste generated from seafood processing industry, including various by-product recovery options. Role of waste minimization and cost effectiveness in sustainable seafood waste management was also discussed. Pre-treatment opportunities for effluent and water conservation are primary targets for pollution prevention practices in seafood processing industries.

In **Chapter 3**, the results of the survey conducted in ASIB on seafood waste management status and management and pollution issues were analyzed in detail. The study period was divided into, pre trawl ban period from January to May, trawl ban period from June to July and post trawl ban period from August to December. The primary data regarding the waste management status and issues were collected through census method using Semi Structured Interview Schedule (SSIS) questionnaires, specifically prepared for the survey units

such as seafood processing units, pre-processing units, fish markets, and shell waste utilizing units. It was found that the seafood processing units in ASIB are presently working only at 31 % of their total available capacity. The peak season of processing activity was found to be the post trawl ban period from August to December followed by the pre trawl ban period from January to May and minimum during trawl ban months of June and July. The cephalopods, shrimps and finfish formed the major seafood varieties currently being processed in the area. A new trend was identified in tuna being emerging as a major seafood variety in the area forming about 21 % of total procurement annually. In ASIB, among the seafood waste generated by the seafood processing units (22643.4 t/year), the maximum quantity was accounted by the shell waste (10625 t/year) followed by finfish waste (6437 t/year) and cephalopod waste (5581.4 t/year). In the case of seafood pre processing units, only 42.3% are having an Effluent Treatment Plant (ETP) and about 22.7% of the Effluent Treatment Plants installed by the processing units in ASIB are currently operating under capacity, by handling more liquid waste than the designed capacity. The whole amount of solid waste generated in ASIB is disposed off by the private contract system which includes the lesser valorised portion and a greater non-utilized portion. The case study revealed that the seafood pollution not only exerts environmental impacts but also livelihood and health issues among the coastal community. The major issues pointed out by the local community were smell and breathing problems, ground water pollution, skin and stomach diseases, mortality of native fish species, destruction of agricultural crops and clogging of the water canal.

In **Chapter 4**, analysis of the different classes of seafood waste available in the study area for their nutritional parameters was conducted, in order to highlight their nutritive quality and to suggest possible valorisation options. The seafood processing waste samples were collected from various waste generation points in the study area, such as fish markets, pre-processing units, processing units and shell waste utilization units. The samples were analyzed for proximate composition (moisture, protein, fat and ash), amino acid profile, fatty acid profile and chitin content. The proximate analysis inferred the superior quality of the seafood processing waste varieties in the area which are rich in protein (9.5 - 24.5%) and lipid (0.34 - 13.5%) content. The amino acid analysis further proved the quality of the seafood waste protein, highlighting the presence of essential amino acids. The fatty acid profile analysis highlighted the fact that the seafood waste materials are an excellent source of PUFA rich seafood oil which could be industrially utilized for oil extraction. The chitin content of the shell waste (4.4-5.2%) available in the study area was also found to be within the acceptable range for industrial utilization.

In **Chapter 5**, the objective of the study was to identify a possible solution for treating the bulk quantity of seafood waste generated in the area. Production of silage from seafood waste was identified as the most suitable technology because of its low investment and simple technology involved. Further, developing an organic liquid fertilizer from seafood silage for foliar application in plants was suggested. The ingredients and methodology involved in silage production was environmental friendly and 100% within organic certification standards. The

chapter also gives an extensive review on seafood silage production and foliar fertilization. Different foliar formulations were prepared from Tuna Viscera (TV), Fish Waste (FW) and Shrimp Head (SH). The formulations were later subjected to NPK (Nitrogen, Phosphorus and Potassium) analysis, secondary and micronutrient evaluation, evaluation of surface tension and also pH at 2% dilution. The NPK analysis of the seafood silage based foliar preparations showed that the primary nutrients such as Nitrogen, Phosphorus and Potassium were present in comparable ratios to that of a commercial formulation used in the study. The secondary and micronutrient evaluation also revealed that the seafood foliar formulations were comparable to commercial samples and furthermore, some of them such as TV100 showed a better balanced ratio. The analysis of surface tension (ST) of the foliar formulations at 2% level indicated that compared to commercial and control, the seafood formulations had a lower ST (0.021 – 0.026 dynes/cm.) favouring their nutrient absorption rate. The pH of the test formulations were also found to be in the desired range (2.85 – 3.65) for maximum nutrient absorption, cementing their candidature as potential foliar spray formulations.

In Chapter 6 the effect of the seafood silage based organic foliar formulations were demonstrated on two common vegetables, okra (*Abelmoschus esculentus*) and red amaranth (*Amaranthus tricolor L.*). The entire experiment was carried out in a completely randomized design (CRD). The experiments were repeated twice for rectifying the errors and then the final experiment was conducted. The seafood foliar formulations (TV and FW) were applied on plants and the subsequent effects on plant height, leaf area and yield were measured and compared with that of a control, a recommended chemical solid fertilizer and a commercial foliar fertilizer. The hypothesis of impact of seafood pigments on leaf colour was tested on amaranthus using shrimp head silage (SH) prepared from two batches of treated and non treated silages. The chemical and organic soil fertilizer application rates were as per the recommendations of Kerala Agricultural University. The study on okra proved that even at 50% of recommended chemical NPK dosage, it is possible to generate a similar or high yield when complemented with foliar fertilization of TV @ 1% concentration. All the seafood silage based foliar formulations were 47-140% better in terms of yield when compared with commercially available foliar fertilizer. The commercial foliar application, even though generated better plant height and leaf area, could produce only identical yield results with that of the control.

The study on amaranthus indicated that plants treated with only 25% of the recommended chemical NPK dosage when supplemented with tuna viscera foliar spray @ 1% concentration, produced a plant yield which was significantly higher than the 100% NPK application. Seafood foliar spray consisting of shrimp head and tuna viscera treated (75/25) and sprayed @ 1% concentration produced a 52% higher yield than the commercial foliar formulation. The hypothesis on effect of pigment preservation of SH silage on leaf colour enhancement on amaranthus leaves was rejected as, there was no difference in coloration in leaves of plants sprayed with pigment- preserved and unpreserved samples of shrimp head silage- based foliar formulation. The leaf colour analysis indicated that the seafood foliar spray consisting of shrimp head silage and sprayed @ 1% concentration generated a 200% higher redness value (a*) than

the commercial foliar formulation and chemical NPK treated plants. This property was confirmed to be the effect of nitrogen delivery efficiency of seafood foliar sprays.

Even though higher in terms of input and labour costs when compared to the heavily subsidised chemical fertilizer cost ,causing environmental and economical ill effects by the unregulated usage chemical fertilizers, it could be recommended that the seafood foliar fertilization, in the long run, may be adopted as a reliable organic agricultural input.

Concept of sustainable seafood waste management

From the entire study, a concept for the sustainable management of seafood waste in ASIB is recommended, which could be adopted in a long run for maintaining the flow of raw materials, products and waste within the cycle of sustainable utilization and management .The concept consists of channelizing the solid seafood waste for the production of seafood silage, which could be further utilized for the production of organic liquid fertilizer and livestock feed, other than the major share of shell waste being utilized for chitin/chitosan preparation. The liquid waste generated has to be subjected to efficient treatment using energy efficient and low cost technologies available before discharging to the immediate water ecosystem. The livestock feed and liquid fertilizer generated could create additional job opportunities to the local residents, as small scale industrial ventures, providing income especially to women groups. This will contribute towards the livestock rearing sector and organic farming sector in the form of low cost, nutritionally superior feed and reliable organic fertilizer. The output generated from livestock in the form of meat, milk and egg and that from organic farming, in the form of healthy vegetables, would again enter the food chain, creating a sustainable seafood waste fuelled integrated system.

Future perspectives

- Field level application of seafood silage foliar formulations should be conducted in collaboration with agricultural research stations to study their effect.
- The minimum frequency of application, without compromising on the yield, needs to be standardized for optimizing the labour input costs.
- Further studies need to be conducted on high value organic crops such as tea, spices and fruits for assessing the effect of seafood foliar spray application.
- Application of seafood foliar formulations along with drip irrigation water to be effective as a slow release nutrient and as a liquid soil conditioner could be investigated.
- Seafood foliar formulations could be applied in futuristic agriculture methods such as poly house farming, hydroponics, airoponics and aquaponics as an organic fertilizer input.



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APPENDIX 1

ANEXURE 3.1

QUESTIONARE

Fish Processing Units

Name of the firm :
 Name :
 Age :
 Gender : M F
 Occupation :
 Year of starting :
 Total capacity :
 Number of working days in a year :
 Kind of approval of the plant :

Q1	What are the varieties of raw materials used?		
Q2	What is the average quantity of different raw materials produced per day?		
	January-May	June-July	August-December
Q3	What is the average quantity of waste generated per kg for each of the variety of raw materials processed?		
Q4	What is the average quantity of water used for fish processing per day?		
Q5	Is there any waste treatment facility in the plant? Yes/No. If yes, specify.		

Q6	If there is no waste treatment facility in the plant, What is the present method of waste disposal followed?
Q7	Do you have to incur any expenditure for disposing waste? Yes/No. If yes, how much?
Q8	What are the problems faced related to the fish waste management?
	1. 2. 3.
Q9	Are you aware of methods of fish waste utilization? Yes/No. If yes, specify.
Q10	If you know about fish waste utilization, what are the problems to practice it?
Q11	Any suggestions for better waste management?
Q12	If a better arrangement of fish waste utilization is made, you prefer to do it in your plant or get done by an out side Agency?

ANEXURE 3.2

QUESTIONARE

Fish Processing Units/Pre-processing Units/Shell Waste Utilizing Units

WASTE MANAGEMENT

Q1	What is the average quantity of solid waste generated per day?
Q2	What is the average quantity of liquid waste generated per day?
Q3	What is the management practice for solid waste?
Q4	What is the management practice for solid waste?
Q5	Is there any Effluent treatment plant (ETP), Biogas plant (BP) or any other waste treatment facility in the plant? (YES/NO) Please specify
Q6	What is the initial cost of investment for the current waste treatment facility?
Q7	What is the daily operational cost for the current waste treatment facility?
Q8	What is the technology used for the current waste treatment facility? (How does it work?)
Q9	From where the technology was transferred?
Q10	What is the working efficiency of the current waste treatment facility?
	Efficient <input type="checkbox"/> Inefficient <input type="checkbox"/>
Q11	If it is inefficient, what may be the reason?
Q12	What is the total capacity of the current waste treatment facility?

ANEXURE 3.3
QUESTIONARE
Fish Pre-Processing Units

Name :
 Age :
 Gender : M F
 Occupation :
 Educational qualification :
 Number of working days in a year :
 Year of starting :

Q1	What are the varieties of raw materials used?		
Q2	What is the average quantity of different raw materials used per day?		
	January-May	June-July	August-December
Q3	What is the average quantity of waste generated per kg for each of the variety of raw materials processed?		
Q4	What is the average quantity of water used/day by the plant?		
Q5	What is the present method of waste disposal followed?		
Q6	Do you have to incur any expenditure for disposing waste? Yes/No. If yes, how much?		
Q7	What are the problems faced related to the fish waste management?		

	1.
	2.
	3.
Q8	Are you aware of methods of fish waste utilization? Yes/No. If yes, specify.
Q9	If you know about fish waste utilization, what are the problems to practice it?
Q10	Any suggestions for the improvement of present method of waste treatment?
Q11	If a better arrangement of fish waste utilization is made, you prefer to do it in your plant or get done by an outside Agency?

ANEXURE 3.4
QUESTIONNAIRE
Fish Markets

Name :
 Age :
 Gender : M F
 Occupation :
 Educational qualification :
 Number of working days in a year :
 Year of starting :

Q1	What is the average quantity of different raw materials produced per day?		
	January-May	June-July	August-December
Q2	What is the average quantity of waste generated per day?		
Q3	Is there any fish loss as waste other than cleaning and cutting waste?		
Q4	What is the present method of waste disposal followed?		
Q5	Do you have to incur any expenditure for disposing waste? Yes/No. If yes, how much?		
Q6	What are the problems faced related to the fish waste disposal?		
	1.		
	2.		
	3.		

Q7	Are you aware of methods of fish waste utilization? Yes/No. If yes, specify.
Q8	If you know about fish waste utilization, what are the problems to practice it?
Q9	Any suggestions for the improvement of present method of waste disposal?

ANEXURE 3.5

QUESTIONARE

Shell Waste Utilizing Units

Name of the firm :
 Name :
 Age :
 Gender : M F
 Occupation :
 Year of starting :
 Total capacity :
 Number of working days in a year :
 Kind of approval of the plant :

Q1	What are the varieties of shell waste procured per day?		
Q2	What is the average quantity of shell waste derivatives produced per day?		
	January-May	June-July	August-December
Q3	What is the average quantity of solid waste generated per day?		
Q4	What is the average quantity of water used used for processing per day?		
Q5	What is the average quantity of liquid waste generated per day?		
Q6	Is there any waste treatment facility in the plant? Yes/No. If yes, specify.		

Q7	If there is no waste treatment facility in the plant, What is the present method of waste disposal followed?
Q8	Do you have to incur any expenditure for disposing waste? Yes/No. If yes, how much?
Q9	What are the problems faced related to the fish waste management?
	1. 2. 3.
Q10	Are you aware of methods of fish waste utilization? Yes/No. If yes, specify.
Q11	If you know about fish waste utilization, what are the problems to practice it?
Q12	Any suggestions for better waste management?
Q13	If a better arrangement of fish waste utilization is made, you prefer to do it in your plant or get done by an out side Agency?

ANEXURE 3.6 QUESTIONNAIRE

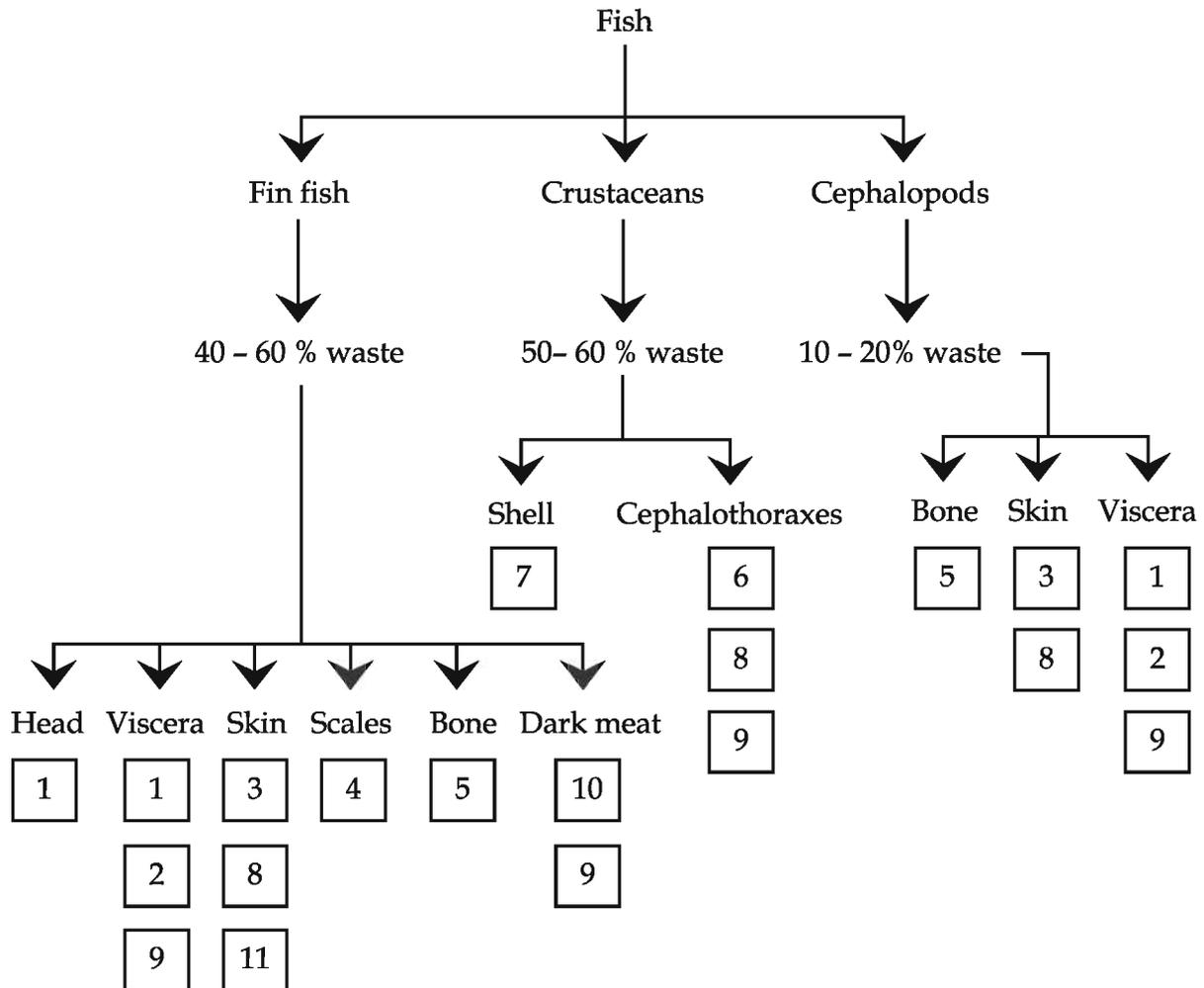
Residents

Name of the firm :
 Name :
 Age :
 Gender : M F
 Occupation :
 Year as resident of the area :

Q1	Is there any problem in the area related to fish waste disposal? Yes/No. If yes, specify.
	1. 2. 3.
Q2	When did the problem due to fish waste started in the area?
Q3	Any agitation has occurred against fish waste problem in the area? Yes/No. If yes, specify.
Q4	What was the result of the agitation?
Q5	Which are the agencies involved in reacting to waste problem?
Q6	Any suggestion for solving the present problem?
	1. 2. 3.

APPENDIX 2

Waste Utilisation Index (WUI) for seafood processing waste available at ASIB



- | | | |
|-----------------------------------------------------------------------------|---------------------------------------------------------------------------------------|-----------------------------------------------------------------------------|
| 1 Silage | 5 Bone / Cartilage powder | 9 Fish oil |
| 2 Enzyme | 6 Extracts | 10 Protein |
| 3 Collagen | 7 Shell derivatives | 11 Fish leather |
| 4 Pearl essence | 8 Pigments | |

APPENDIX 3
The SAS system
The GLM Procedure Okra

Table 6.1 Dependent Variable: plheight

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Ph covariant	1	863.05587	863.05587	2.47	0.1272
Treatment	9	10441.04645	1160.11627	3.31	0.0067
Error	29	10152.44413	350.08428		
Total	39	22322.40000			

Table 6.2 Dependent Variable: leaf area

Source	DF	Type III SS	Mean Square	F Value	Pr > F
La covariant	1	11.0769	11.0769	81.88	<.0001
Treatment	9	102549.3967	11394.3774	84229.0	<.0001
Error	29	3.9231	0.1353		
Total	39	316171.1000			

Table 6.3 Dependent Variable: yield

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Treatment	9	116648.4000	12960.9333	24301.8	<.0001
Error	30	16.0000	0.5333		
Corrected Total	39	116664.4000			

The GLM Procedure Amaranthus

Table 6.4: Dependent Variable: Plant height

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Ph covariance	1	48.00531915	48.00531915	2836.43	<.0001
Treatment	14	72.60462452	5.18604461	306.42	<.0001
Error	44	0.744681	0.016925		
Corrected Total	59	1084.194000			

Table 6.5: Dependent Variable: Leaf area

Source	DF	Type III SS	Mean Square	F Value	Pr > F
La covariance	1	48.75000	48.75000	Infty	<.0001
Treatment	14	27250.54885	1946.46778	Infty	<.0001
Error	44	0.00000	0.00000		
Corrected Total	59	38741.14653			

Table 6.6: Dependent Variable: Yeild

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Treat	14	214511.1136	15322.2224	14143.6	<.0001
Error	45	48.7500	1.0833		
Corrected Total	59	214559.8636			

The GLM Procedure *Amaranthus* Leaf Color

Table 6.7: Dependent Variable: L*

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Treatment	14	110.0861867	7.8632990	3.30	0.0029
Error	30	71.4039333	2.3801311		
Corrected Total	44	181.4901200			

Table 6.8: Dependent Variable: a*

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Treatment	14	225.3608311	16.0972022	36.46	<.0001
Error	30	13.2461333	0.4415378		
Corrected Total	44	238.6069644			

Table 6.9: Dependent Variable: b*

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Treatment	14	360.3164667	25.7368905	17.28	<.0001
Error	30	44.6769333	1.4892311		
Corrected Total	44	404.9934000			

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2. **Sasidharan, A.,** Baiju, K.K. and Mathew, S., 2010. Current status of seafood processing waste management and its impact on coastal community in Cochin Corporation, India. *Int. J. of Environ. and Waste Manage.* 12(4), 422-441.
3. **Sasidharan, A.** and Mathew, S., 2009. Seafood waste management in aroor seafood industrial belt: Status, impact and opportunities, In: Coastal Fishery Resources of India: Conservation and Sustainable Utilization (Meenakumari, B., Boopendranath, M.R., Edwin, L., Sankar, T.V., Gopal, N. and Ninan, G., Eds.), 693-702.



THE HINDU

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Fish waste as plant supplement

K.S. Sudhi

Photo: Vipinchandran



New finding: Researchers spraying locally-developed 'foliar sprays' on ladies finger plants at a laboratory of the School of Industrial Fisheries of the Cochin University of Science and Technology. --

KOCHI: Plant growth stimulants from fish waste — it's the new organic way of farming being attempted at a lab in Kochi. A researcher at the School of Industrial Fisheries of the Cochin University of Science and Technology has offered to give 'foliar sprays' developed from fish waste. Management of fish waste has been difficult in areas like the Aroor industrial belt and Kochi Corporation. Waste is found dumped on vacant plots or nearby water bodies. This has led to outbreak of skin diseases and other health hazards in Chandiroor and its adjoining areas. It is estimated that the fish processing plants located at Aroor generate 128 tonnes of waste every day, and Kochi adds another 48.14 tonnes. "The attempt was to create a cost-effective and eco-friendly way to manage the fish waste, which has been causing serious health issues," said S. Abhilash, Principal Investigator of the project. The Science and Society Division of the Department of Science & Technology, New Delhi, has supported the project. Foliar sprays are micronutrient mixtures sprayed on plants to accelerate their growth. The micronutrients are directly absorbed by the plant through the specialised cells under the leaves. Foliar spray is developed from fish silage, the liquefied fish protein made by adding formic acid to fish parts. The spray can be marketed and promoted as part organic farming, especially among gardening enthusiasts, said Mr. Abhilash. Cattle feed can also be developed from the fish silage. Though some companies are marketing foliar sprays at Rs. 80 for 100 grams, the one developed by the lab could be sold at half the price, he said. The researchers of the University are trying the spray on ladies finger plants farmed on the campus. They have also trained around 100 women in Kuthiyathode for the production of sprays and cattle feed. Attempts are also made to create a marketing network for the products.

<http://www.hindu.com/2009/07/04/stories/2009070455750900.htm>

THE HINDU

Online edition of India's National Newspaper

Wednesday, Mar 02, 2011

ePaper | Mobile/PDA Version

Turning fish waste into products

Staff Reporter

KOCHI: Ever thought about converting fish waste in to value-added byproducts?

A young researcher at the School of Industrial Fisheries in Cochin University of Science and Technology is earnestly pursuing a project that would explore the immense possibilities of fish waste management for the welfare of the society. Abhilash S., Principal Investigator of the project, has developed a particular foliar spray (organic plant nutrient supplement) combining different fish wastes at a particular combination and following a particular protocol and concentration.

"It was tested on Red Amaranthus (red cheera) and confirmed boost in growth and colour," he said.

Mr. Abhilash, who is planning to file a patent for this formulation, is now studying its potential in Hydroponics and as an aquarium plant nutrient.

"My observations confirmed that the usage of fish waste based foliar spray in farming could reduce the cost considerably. Plants taken for the study had given around 20 per cent more yield when it were supplemented with the formulation developed as part of the research.

The recommended dose of chemical fertilizers like urea, potash and phosphate was also reduced by 75 per cent (in the case of Red Amaranthus)," he said.

Mr. Abhilash said that the fish waste based foliar sprays could be priced at half the rate compared to the existing organic foliar sprays that cost around Rs. 80 - 100 per 100 ml.

Byproducts

The researcher had the opportunity to carry out a study on fish waste management and its socio-environmental impact assessment in Kochi Corporation and Aroor industrial areas under the young scientist scheme of the Department of Science and Technology.

"It was observed during the study that about 400 to 500 tonnes of solid waste and around 8 to 10 lakh litres of liquid waste were generated by the seafood processing and pre-processing units in both Kochi and Aroor areas on a daily basis. Only 25 to 30 per cent is currently being utilised for byproduct development in the case of solid wastes," he said. Mr. Abhilash said that the development of a fish waste management system based on local participation would benefit the industry and the community in a big way.

The industry could reduce waste management issues while the community would be able to generate job and income opportunities through production and marketing of value added products from fish waste, he said.

<http://www.hindu.com/2011/03/02/stories/2011030250730200.htm>



FACULTY OF MARINE SCIENCES

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