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From the Editorial Board.....

In this issue of FishTech Reporter, we are happy to introduce a break-through research finding. Bacterial aetiology in shrimp post-larvae infection is a crucial milestone in combating mass mortality of shrimps. This has been documented in our report. Food loss from gillnets due to depredation, confirmed through underwater observation is the first report of its kind in the country. Seaweeds, being the richest sources of minerals among all plant resources; this edition brings out the elemental profile of most common seaweeds along the Indian coast and infers their suitability for human consumption. Effective utilization of fish processing discards is essential for better value realization and for reducing environmental pollution. We discuss the advantages of converting fish roe mass into functionally stabilized fish roe powder and protein hydrolysates from jawala shrimp—using proteolitic enzymes. This issue also dwells into an innovative approach to combat marine fouling on cage netting by applying nanomaterials. The article on practical application of slurry icing system against conventional icing system for onboard preservation of fish is of special interest. Pre-osmotic blanching, a simple technology for value addition of small sized squids is also covered in this issue. Articles on trap fishing show the shifting focus towards low environment impact fishing gears. We are happy to bring out these knowledge inputs for the understanding of the stakeholders we serve.

Mass mortality of *Penaeus monodon* post-larvae due to *Vibrio cholerae* O139 infection

Toms C. Joseph, Murugadas V. and Lalitha K.V.

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Vibriosis is one of the most serious threats in the commercial production of larval penaeid shrimp. Large-scale mortalities in larval penaeids associated with Vibrio harveyi have been observed globally (Lightner, 1996). Disease outbreaks attributed to other Vibrio species such as V. alginolyticus, V. damsela, V. parahaemolyticus, V. vulnificus and V. penaeicida have been reported in nursery or growout ponds of *Penaeus vannamei*, P. monodon, P. japonicus and P. stylirostris worldwide (Brock and Lightner, 1990; Ishimaru et al., 1995). V. anguillarum, V. campbelli, V. nereis, V. cholerae (non 01) and V. splendidus have also been reported in association with disease outbreaks in shrimps (Chen 1992; Lavilla et al., 1990; Esteve and Quijada, 1993; Sahul-Hameed et al., 1996). Vibriosis in penaeid shrimp post-larvae might be due to either opportunistic Vibrio flora or to pathogenic Vibrio spp. specifically infective at one or more larval developmental stages.

In this study, we investigated the cause of mass mortality in *Penaeus monodon* post-larvae in farms located in Ernakulam, Kerala (India). The symptoms of the disease included lethargy and reddish discolouration of the affected post-larvae. The infected P. monodon post-larvae died within 48 h after the infection. The infected post-larvae were tested for pathogenic bacteria by plating on to Tryptone Soy Agar (TSA) and Thiosulphate Citrate Bile Salts (TCBS) Agar. Bacterial isolates obtained from the moribund and dead shrimp were confirmed as belonging to V. cholerae O139 sero group, the aetiological agent of cholera in humans, using O139 serogroup-specific antiserum and by a PCR based assay targeting rfb-O139 gene (Fig. 1). The isolates were found to carry cholera toxin gene ctx and genes coding for virulence determinants; zot and tcpA in PCR assay.

Vibrio cholerae is an autochthonous flora of brackish water and estuarine systems. V. cholerae O1 and O139 are the major serogroups that cause outbreaks of cholera in human beings. More than 200 sero groups of V. cholerae has been identified so far and epidemic cholera has been confined only to isolate within serogroups of O1 and O139. The cholera toxins produced by V. cholerae are the causative agents of cholera. There are only limited reports of infections in post-larvae of shrimp due to V. cholerae. A V. cholerae non-O1, non-O139 isolate from a shrimp farm was reported to be pathogenic to post-larvae and juveniles of P. monodon larvae (Halder et al., 2007). In this study, a highly virulent V. cholerae O139 strain was isolated from infected shrimp post-larvae.

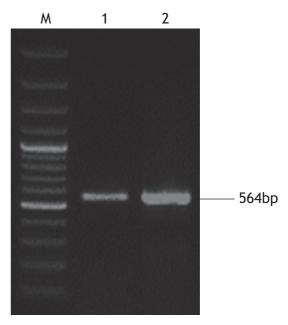
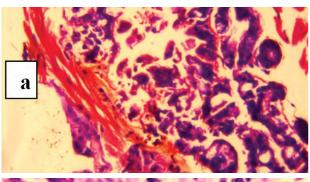


Fig. 1. Detection of *V. cholerae* O139 cholera toxin gene by PCR. Lane 1 - *V. cholerae* MTCC3906; Lane 2 - Isolated strain from infected post-larvae; Lane M - 100bp plus ladder (MBI Fermentas, US)

Experimentally exposed shrimp post-larvae with V. cholerae O139 exhibited significant mortalities that increased with increasing doses of bacteria. The LD₅₀ value of one of the isolates was determined in post-larvae of P. monodon, Fenneropenaeus indicus and Litopenaeus vannamei which ranged from 4.6x104 for L. vannamei to 7.1x106 for P. monodon. V. cholerae was reisolated from experimentally infected moribund shrimps. Histopathological examination revealed the presence of large numbers of bacteria laden in the hepatopancreas of the infected postlarvae. There was rupture of basal laminae of the hepatopancreatic tubules and severe necrosis, loss of structure, atrophy, vacuolation and rounding into the lumen of tubular epithelial cells, which suggest that tissue integrity was affected in shrimp due to the infection (Fig.2). The bacterial strain isolated from moribund P. monodon post-larvae was identified as the causative agent of the mortality by isolation, subculture, reinfection and reisolation according to Koch's Postulates. To our knowledge, this is the first report of V. cholerae O139 strain causing high mortalities in shrimp.



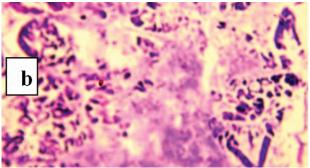


Fig. 2(a) Healthy hepatopancreas in control larvae. (b) Pathology of hepatopancreas in *P. monodon* larvae showing severe necrosis, rounding and sloughing of cells, when infected with *V. cholerae*

This study has wide repercussions since the infected post-larvae can serve as a host for

multiplication and spread of the highly pathogenic *V. cholerae* O139 outside the human host. The involvement of cholera toxin and other regulatory genes in the pathogenesis of shrimp need to be ascertained.

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Functional and physico-chemical properties of spray dried rohu roe powder stabilized with gum arabic

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Fish roe is a very popular protein source throughout the world with premium prices for selected roe products such as caviar from surgeon fish and salmon. Roe from rohu fish (Labeo rohita) is an abundant and under-utilized byproduct. In general, fish roes have a favorable essential /nonessential amino acid ratio (0.93:1.23), and are considered as a food source of high-quality protein. Apart from that, it has high amount of unsaturated fatty acids (MUFA and PUFA), hence it can serve as a valid nutritional supplement. Rohu roe contains 23.6% n-3 fatty acids and EPA (20:5), DHA (22:6) in the range of 1.5% and 11.8%, respectively. Direct use of fish roe for food formulation has its own implications as its addition may decrease sensory acceptance of the food due to intense flavour and odour of the roe as well as the oxidation of unsaturated omega-3 fatty acids. Microencapsulation and spray drying is commonly employed in food industry to stabilize fish oil against oxidation by enclosing the oil in a hard shell matrix of suitable polymers. Once the fish roe is micro-encapsulated, the resulting powder can be added to a wide range of foods without affecting the palatability of the product. Products fortified in this way are considered to be more bio-available and have the capacity to increase the intake of PUFA especially EPA and DHA.

In the present study, spray dried powder was prepared from roe of *Labeo rohita* and the physicochemical and functional properties were evaluated. The roe emulsion was stabilized with gum arabic prior to spray drying. The spray dried powder was analyzed for its proximate composition, physico-chemical and surface-active properties, morphological characteristics, amino acid composition, fatty acid composition, particle size analysis and zeta potential, gastro-intestinal digestion and release pattern.

Proximate composition of fresh roe, spray dried fish roe and fish roe with gum arabic is

presented in Table 1.

Table 1. Proximate composition of fresh and spray dried rohu roe

	Mean Value (%)			
Parameters	Fresh roe	Spray dried	Fish roe with	
		fish roe	gum arabic	
Moisture	70.10	6.23	5.65	
Fat	20.42	12.34	11.08	
Ash	3.92	4.64	4.89	
Total crude protein	70.69	71.43	73.36	

The spray dried powder exhibited more than 90% solubility in distilled water. The isoelectric point of the spray dried powder was found to be at pH 5.5-6. Incorporation of gum arabic improved viscosity, emulsion capacity and fat binding properties of the reconstituted powder. The surface morphology of the powder as revealed by Scanning Electron Microscopy indicated smooth and wrinkle free surface for gum-stabilized sample (Fig. 1). The spray-dried powders were further characterized for particle size and zeta potential by dynamic light scattering method. The particle size analysis indicated presence of peaks at two different size ranges for gum stabilized powder, the larger ones being the encapsulated roe particles; whereas control fish roe powder had a single peak of medium dimension indicating the predominance of unencapsulated particles (Fig. 2). Incorporation of gum arabic improved the shelf life of roe powder in terms of reduced selfaggregation and fat oxidation reactions as indicated by free fatty acid profile. The results of the present study suggest that converting the fresh soft textured raw mass into more stable fish roe powder with good nutritive value as well as functional properties widens the scope for utilization of this under-exploited food resource.

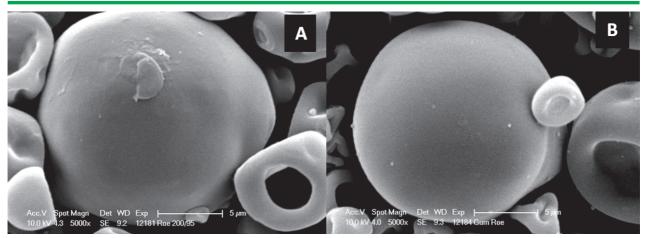
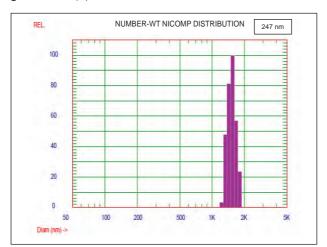


Fig. 1. Scanning electron microscopy images of the fish roe spray-dried powder (A) and fish roe spray dried powder with gum arabic (B)



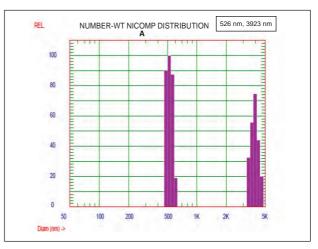


Fig. 2. Particle size distribution of spray dried fish roe powder (A) and fish roe spray dried powder with gum arabic (B)

Spray drying of fish roe with gum arabic is a novel method for utilizing fish roe discarded during the pre-processing stage. Besides by preservation and value addition of fish roe by this method, a valuable nutritional supplement can be made available.

Radical scavenging activity of protein hydrolysates of *Acetes indicus* derived using Protamex® and Bromelain

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Generation of immense quantity of under-utilized marine bycatch from commercial fisheries causes not only environmental pollution but also problems like foul smell, proliferation of insets and pests etc. If not treated, the recoverable components

from this valuable biomass will be lost. Thus, in recent years, marine bycatch has attained a greater importance as a source of various bioactive compounds.

Acetes indicus (Jawala Shrimp), a shrimp species, abundant in Veraval coast of Gujarat are caught in trawl net as bycatch. It is used mainly for fish meal preparation and is distributed across the country. In 2013-14, the landing of nonpenaeid shrimp was 107122.77 t including 94 962.78 t of Acetes in Gujarat coast (CMFRI, 2014). Since not preferred for human consumption, it is under-utilized and has low commercial value. In crustaceans and molluscs, protein levels vary from 7 to 23% (w/w) (Murray and Burt, 2001). The proximate composition reveals that it contains a high amount of protein and it is a valuable raw material for further processing. Therefore, it is worthwhile to use jawala shrimp as a raw material for the preparation of protein-based products.

The promising bio-technique which is currently employed to recover the nutritionally and physiologically important peptides is enzymatic hydrolysis. The hydrolysis converts the fish proteins to fish protein hydrolysates (FPH) which contain biologically active peptides. Proteases used for hydrolysis is one of the key critical process parameters for the production of hydrolysates with improved bioactive properties. Although numerous bioactive peptides have been reported from fish sources, studies on jawala shrimp are limited. So the present investigation was aimed to study the antioxidant activity of hydrolysate from jawala shrimp as influenced by specific enzymes.

The *Acetes* sp. was purchased from market under iced condition. The samples were stored at -20 °C for further study. The proximate composition (moisture, protein, lipid and ash) was determined as per AOAC (2000) which showed that *Acetes* sp. contained 86.80% moisture, 8.12% crude protein, 1.54% crude fat and 2.52% ash.

Optimum conditions for hydrolysis, which included enzyme/substrate ratio and time of hydrolysis were standardized. Two proteolitic enzymes, Bromelain and Protamex® were evaluated for hydrolysis of the shrimp protein. Hydrolysates were prepared according to the method described by Benjakul and Morrissey (1997). Reaction mixture was incubated at 50 °C, for different duration viz., 5, 10, 20, 30, 40, 50 and 60 min. for hydrolysis. Lowry method was used for protein estimation. Antioxidant activity of the

hydrolysates was determined in terms of DPPH free radical-scavenging activity (Blois, 1958).

In the present investigation, hydrolysate produced using Protamex® was found to have higher scavenging ability (67.8%) than the hydrolysate produced by Bromelain (56.5%) (Fig. 1). Protein hydrolysates differ in their antioxidant properties based on the nature of proteases used as the sequence of released peptides differ chiefly based on the specific activity of the enzyme towards the substrate (Elavarasan *et al.*, 2012).

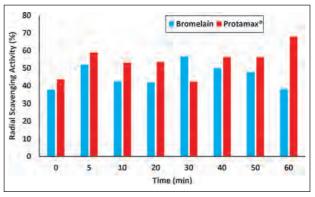


Fig. 1. DPPH radical scavenging activity of protein hydrolysate from *Acetes indicus*

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Quality evaluation of croaker fish (Johnius dussumieri) under slurry ice

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Freshness of fish is an important factor which determines its commercial value and potential for export. One of the main pre-occupations of seafood industry is to improve or maintain the quality of perishable foods to reach a final product with optimal quality. Among various methods currently used, the most important are those based on the application of low temperatures that preserve taste and nutritional value of fresh material. In the last couple of decades, normal ice was indispensible for preservation of fish. Though, refrigerated seawater and addition of chemical preservation agents have been used for preservation of fish species, till date there has been no suitable alternative for normal ice. Slurry ice has been reported to be a promising technique for the preservation of aquatic food products in an ice-water suspension at sub-zero temperature (Huidobro et al., 2000). It is a biphasic system consisting of small spherical ice crystals surrounded by seawater at sub-zero temperature. Two relevant characteristics of slurry ice are, its faster chilling rate, which is a consequence of its higher heat-exchange capacity; and the reduced physical damage caused to food products due to its microscopic spherical particles as compared to the damage elicited by flake ice. In addition,

complete coverage of the fish surface by the slurry ice mixture minimizes surface dehydration. Although the theoretical advantages of slurry ice are well known, few empirical data have been reported about the potential practical application of slurry ice for preservation of fish compared to normal ice.

In the present study, the effects of slurry ice on quality changes of dhoma (*Johnius dussumieri*) fish were evaluated and it was compared with the traditional flake ice. The slurry ice was obtained from the M/s Chirag Ice International, Navi Mumbai and the fishes were submerged in it (Fig 1). Similarly, another set of fishes was kept in normal flake ice as control. Microbiological and biochemical changes of fishes under both the treatments were assessed. Biochemical parameters considered included proximate composition, TMA, TVB, alpha amino nitrogen, salt soluble nitrogen peroxide value and thiobarbituric acid value. Proximate composition showed that dhoma fish contained 79.87% moisture, 17.38% protein, 0.53% fat and 1.20% ash. Moisture content of the fish samples in slurry ice and control showed increasing trend (78-83%) during storage. However, there was no significant difference (p<0.05) in moisture content between the samples. During storage FFA, PV and TBA values of the samples



Fig. 1. Fishes submerged in normal flake ice and slurry ice

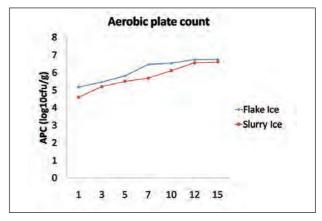


Fig. 2. Changes in aerobic plate count during storage

also showed an increasing trend while texture profile analysis showed a decreasing trend. However, sample kept under slurry ice showed better texture (35.40N-28.12N) than conventional ice (27.39N-23.58 N). Colour analysis showed a decreasing trend of L^* value of 68.35-63.37 for fishes stored under conventional ice and 65.18-59.33 for fishes stored under slurry ice during storage period. Microbiological analysis revealed that fishes kept under slurry ice contained lower aerobic plate count (APC), psychrophilic count, *Pseudomonas* sp. and *Brochothrix thermosphacta* count compared to that of fishes stored under flake ice. There was significant difference (p<0.05) in aerobic plate count between the samples during

storage (Fig. 2). The initial aerobic plate count (APC) was 1.6 x 10⁴ cfu/g and it increased to 4.0 x 10⁶ cfu/g and 5.72 x 10⁶ cfu/g for samples kept under flake ice and slurry ice respectively on 15th day of storage. Sensory evaluation revealed that fishes stored in flake ice were acceptable up to 10 days without any change in sensory characteristics, whereas fishes stored under slurry ice had a shelf life of up to 15 days. The study indicated that during 15 days of storage, quality deterioration of fish stored in conventional ice was more pronounced compared to fishes that had been kept under slurry ice. But, oxidative changes were more prominent in the fishes stored under slurry ice.

Pre-osmotic blanching treatment for drying squid rings

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Squid is a highly sought after seafood commodity in the domestic as well as export market because of its high nutritive value. India exported 69569 ton of frozen squid worth ₹ 1275 crores in 2014-15. However, the shelf life of fresh squid is limited on account of high moisture content and nonprotein nitrogen content, which accelerates spoilage mechanisms. Preservation of squid by icing or freezing has several disadvantages like drip loss, texture toughness, nutrient loss etc. Drying is one of the common preservation techniques which can preserve the nutritive value of seafood. It has been demonstrated that osmotic dehydration (cold/hot blanching in brine) prior to drying can improve the final quality and speed up the drying process of fruits and vegetables. However, limited work has been done on preosmotic dehydration treatment in fisheries sector. Frozen squid ring is a product style that fetches high value in the export market. Development of dried squid ring can be a promising technology for value addition of small sized squid. In this background, an attempt has been made to

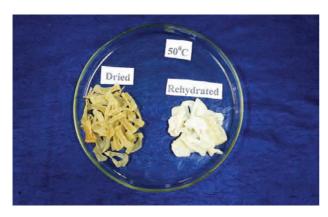
optimize the osmotic blanching necessary to maintain the quality and ring shape of squid rings from small sized squid.

Small sized squids (*Loligo* sp., 100±10g) were purchased from the Visakhapatnam fishing harbour, Visakhapatnam. Rings of 2 cm thickness were cut manually from the de-skinned and cleaned squid. The rings were given blanching treatment at different temperatures *viz*. 50, 55, 60 and 65 °C. The concentration of brine and time of blanching were set as 4% and 10 min., respectively for all the groups. After blanching, the rings were drained and dried at 50 °C in a tray drier to a moisture content of 20-25%.

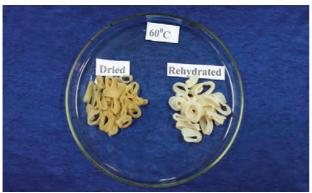
The yield of dried rings from raw squid varied from 18-22%. Temperature of blanching did not influence the yield of final product, whereas it imposed a significant effect on blanching loss. Blanching loss at 50, 55, 60 and 65 °C were 9.5, 11.4, 21-23 and 26-30%, respectively. It was noticed that the drying time required to bring down the moisture content from 80% to

Table 1. Proximate composition of raw squid meat and dried squid rings blanched at different temperatures

Parameter (%)	Raw squid	50 °C	55 °C	60 °C	65 °C
Moisture	80.12	22.95	20.70	23.6	25.42
Protein	16.84	66.47	67.93	64.41	63.54
Fat	3.60	3.14	3.79	3.62	3.71
Ash	2.04	7.06	7.22	7.18	7.84







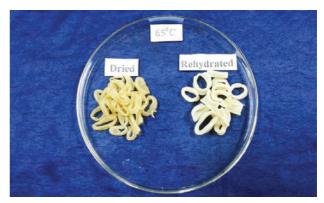


Fig. 1. Appearance of dried and rehydrated squid rings blanched at different temperatures; A) 50 °C, B) 55 °C, C) 60 °C, and D) 65 °C

approximately 20-25% was higher for the rings blanched at 50-55 °C (8-10 h) than those blanched at 60-65 °C (12 to 13 h). Proximate composition indicated meagre difference between the different sample groups (Table 1). The samples dried at 60 and 65 °C were found to be of good quality with respect to its colour and appearance. Blanching at temperatures of 50 and 55 °C failed to obtain a ring shape in the dried as well as rehydrated products (Fig. 1). In addition, significant differences were noticed between the instrumental colour values. Redness (a*) and yellowness (b*) values of the dried squid rings decreased with increasing blanching temperature.

Rehydration curves of different dried samples indicated sharp increase in rehydration rate up to 20 min. followed by a constant rate of rehydration ability. Furthermore, rehydration ability of the samples decreased with increase of blanching temperature. However, the samples blanched at 60 and 65 °C rated higher as per sensory evaluation after cooking (rehydrated for 20 min. and cooking in 1% brine for 10 min.). This study demonstrated that osmotic blanching at 60-65 °C can be used as a pre-treatment to maintain the shape, reduce the drying period and to improve the colour and appearance of dried squid rings.

Elemental composition of Sargassum wightii and Ulva lactuca collected from south east coast of India

Mandakini Devi H., Anuj Kumar, Panda S.K. and Zynudheen A.A.

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Seaweeds are macrophytic marine algae rich in carbohydrates, protein and minerals. These contain elements in higher concentration than terrestrial plants and animal products and some of these elements are essential for our body's physiological functions (Rupe´rez, 2002). Seaweed, a delicacy in Asian countries is consumed in the form of soups and salads. In the present study, major trace elements and heavy metals in green seaweed (*Ulva lactuca*) and brown seaweed (*Sargassum wightii*) were determined using Inductively Coupled Plasma Optical Emission Spectroscopy (iCAP 6300 Duo, Thermo Fisher

Scientific, Cambridge, England) with dual configuration (axial and radial) and iTEVA (version 2.8.0.97) operational software. Mercury analysis was performed by US EPA 7473 method in DMA (Direct Mercury Analyzer)-80 (Milestone, Italy). The seaweed specimens used in the study were collected from Mandapam, Tamil Nadu.

Variation was noticed in elemental composition of both the species. Among the 18 elements analyzed, Ca, K and Na were abundant in S. wightii, while Mg, Ca and Fe were most abundant in U. lactuca (Table 1). Na: K molar ratio of Ulva lactuca (2.03) was higher than S. wightii

Table 1. Mineral composition of Sargassum wightii and Ulva lactuca expressed as mg/100g.

Element	Wavelength (nm)	Sargassum wightii	Ulva lactuca
Ca	422.6	2261.46-2288.57	574.6-558.1
K	766.4	1517.90-1519.76	77.32-77.67
Na	589.5	1309.04-1320.67	92.20-93.63
Ba	455.4	0.10-0.12	1.40-1.46
P	213.6	770.95-782.44	68.85-69.70
Mg	280.2	830.24-855.67	825.8-842.4
Ni	231.6	0.12-0.13	0.29-0.30
As	193.7	1.49-1.62	0.23-0.29
Hg*	-	0.003	0.006-0.007
Pb	220.3	0.37-0.38	ND
Cr	283.5	0.04-0.11	0.32-0.34
Cd	259.9	0.02-0.06	ND
Cu	324.7	0.23-0.27	2.05-2.08
Fe	259.9	17.02-17.56	163.80-167.80
Se	196.0	1.10-1.40	0.28-0.29
Zn	213.8	2.81-2.83	10.06-11.75
Mn	257.6	5.13-5.29	7.33-7.44
Sr	407.7	0.52-0.57	11.39-11.51
Na/K molar ratios**		1.46	2.03

Data presented as range (min.- max.)* Total Mercury analyzed by DMA 80; ** based on mean values; ND: Below the detectable limit

(1.46). Consumption of too much Na and not enough K contributes to hypertension, but both the seaweeds were having balanced Na:K molar ratios. Iron is a vital constituent of haemoglobin. The Fe concentrations in *Ulva lactuca* samples were higher than in brown seaweed. Thus, Ulva could cater to daily intake requirement of Fe for adults i.e. 10-18 mg (Indegaard and Ostgaard, 1991). The presence of elements such as As, Cd, Hg and Pb in food is of concern due to their toxic potential. The seaweeds analyzed didn't have toxic elements in excess level. The concentrations of mercury were less than the FDA's allowable level of 1.0 ppm. Lead and cadmium were not detected in *U. lactuca*. Although total Arsenic was detected at more than 1 ppm level in S. wightii, it is known that only inorganic As is toxic and brown seaweeds are known to contain high amounts of organic Arsenic (Francesconi and Kuehnelt, 2002).

Trace elements like manganese, copper and zinc, considered as essential micro-nutrients were present in very small quantities.

Thus, both the species could very well serve as important source of minerals for our body in the form of food supplements if levels of toxic elements particularly Arsenic is taken care of. This can easily help in meeting the recommended daily intakes of various macro-minerals and trace elements.

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Effect of vacuum packaging on shelf life of monosex tilapia during ice storage

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Relatively few studies have been conducted on the quality and shelf life of freshwater fishes in storage conditions in contrast to the information available for marine species. Among the freshwater fish candidates, tilapia species has emerged from mere obscurity to one of the most productive and internationally traded food fish in the world, to such an extent that they have been called the 'fish of the future'. Common tilapia however has a few disadvantages due to its prolific breeding habit which can be overcome by the production of monosex tilapia, using techniques like hybridization or hormone application.

Packaging makes food more convenient and gives assured safety from biological and chemical changes so that the packaged foods have a longer shelf life. Among the food packaging technologies

developed, vacuum and modified atmosphere packaging has led to the evolution of fresh and minimally processed food preservation, especially in meat and fishery products for the past two decades. The spoilage of fish due to growth and proliferation of aerobic spoilage bacteria and the oxidation of lipids and pigments can be reduced by using the vacuum packaging technique which is a type of modified atmospheric packaging in which, the air present in the pack is completely evacuated by applying vacuum.

Studies on the quality and shelf life of monosex tilapia have not been reported earlier and the potential for utilizing this species for various processing and value addition is enormous. Hence, a study was conducted with the objective of determining the effect of vacuum packaging

on the shelf life extension of monosex tilapia stored under iced condition by evaluating certain physical, chemical, microbiological and sensory parameters. Fresh monosex tilapia samples were cut into steaks of thickness 1.8-2 cm. An initial lot was evaluated for its proximate composition. Further, the steaks were divided into two lots; the first lot was kept as control (AP: air packed) and the second lot was vacuum packed (VP) and ice stored.

Proximate composition of tilapia meat indicated a moisture content of 75.69 \pm 0.01%, 17.47 \pm 0.31% crude protein, 5.17 \pm 0.04% fat and 0.99 \pm 0.01% ash content. Moisture content as well as pH value showed a slightly increasing trend throughout the storage period in both the sample lots. Total Plate Count (TPC) values indicated that AP samples were acceptable up to16th day with a count of 6.8 log cycles whereas VP samples were acceptable upto 19th day with a TPC of 6.59 log cycles (Fig. 1).

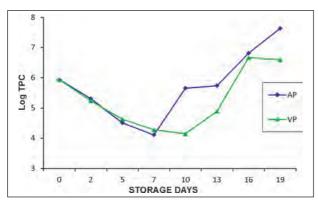


Fig. 1. Variation in Total Plate Count of monosex tilapia steaks during ice storage

An increase in TVB-N was found in AP sample lots compared to VP lots where the TVB-N remained almost constant during storage (Fig. 2). The increase in TVB-N may be attributed to the bacterial decomposition taking place during storage. A TVB-N level of 30-35 mg% is usually regarded as the limit of acceptability (Lakshmanan, 2000) and AP samples crossed the acceptability limit reaching 39.2 mg% towards 16th day whereas VP samples were within the acceptability limit throughout the storage period. TMA-N also increased in both sample lots during storage and AP lot crossed the TMA-N acceptability limit of 10-15 mg N/100 g on 10th day whereas VP

lot remained acceptable till 16th day.

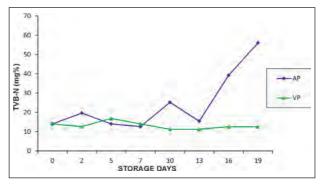


Fig. 2. Variations in TVB-N of monosex tilapia steaks during ice storage

Variations in peroxide value of the sample lots were very low indicating a low level of fat oxidation. Free fatty acid was found to have an increasing trend in all the samples throughout the storage period. TBA value, a major index of fat oxidation was found to increase from an initial value of 0.52 mg malonaldehyde/kg of fish to 1.93 and 0.62 mg malonaldehyde/kg of fish in AP, and VP samples respectively on 23rd day of ice storage which may be on account of the exclusion of air from the latter thereby reducing fat oxidation in the samples.

The air packed samples had better water holding capacity (WHC) than vacuum packed samples. The lightness as well as hardness also decreased for both the sample lots during ice storage. The extent of decrease in lightness was more for VP samples compared to AP samples (Fig. 3).

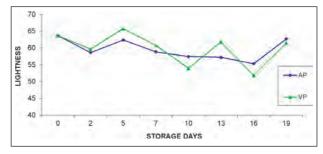


Fig. 3. Variations in lightness values of monosex tilapia steaks during ice storage

Sensory analysis indicated that AP samples were acceptable upto 20th day. The overall acceptance score was below 4.0 (dislike slightly) for VP samples on 22nd day. Based on the sensory and microbiological tests, air packed monosex

tilapia under iced condition, had a shelf life of 16 days whereas in vacuum packed samples, it could be extended to three more days. Hence, from the

present study it is concluded that vacuum packing could help in extending the shelf life of ice stored monosex tilapia steaks.

Assessing domestic fish supply chain - A tool for sustainable fisheries development

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The fish supply chain in domestic marketing is unorganized with complex intra and inter-linkages between market functionaries at different stages of the supply chain. The concept of value chain was conceptualized by Porter and defined as 'A full range of activities that are required to bring a product or service from conception, through different phases of production, delivery to final consumers and final disposal after use'. Improvement in supply chain depends on the combined satisfaction of producers, wholesalers, retailers and consumers. Supply chain comprises of production, distribution and marketing stages which varies across regions. It was stated that sustainability gaps can be improved through integrating various stages of supply chain.

In this pilot study, marketing stage in the supply chain was targeted and the efficiency of major market functionaries (wholesalers and retailers) involved was studied for selected markets. Data were collected from 15 wholesalers

and 15 retailers of three fish markets in Ernakulam district. The marketing costs, marketing margin and intra- and inter-market efficiency were studied. Intra and inter-market efficiency is studied to compare the effectiveness between markets and also among the market functionaries in the markets, by using certain criteria *viz.*, information availability, marketing costs and margins. This is useful in assessing the effectiveness of marketing functions, cost of marketing and method of performing the service in a better way.

The fish supply chain for three fish markets revealed that the fish arrivals to these markets were from outside Kerala (Tamil Nadu, Andhra Pradesh, Odisha and Gujarat) besides local harbours and landing centres. The marketing efficiency of intra and inter-fish markets was assessed using Shepherd's Marketing Efficiency Index (MEI). The inter-market efficiency ranged from 3.11 to 3.46 and the intra-market efficiency

Table 1. Marketing costs, margin and efficiency in the selected fish markets

Markets	Purchase price (₹ in lakhs)	Sale price (₹ in lakhs)	Marketing Margin	Marketing cost	Marketing efficiency
Market I (3.46)					
Wholesaler	25.00	36.67	11.67	0.40	0.78
Retailer	48.00	60.00	20.00	0.32	4.87
Market II (3.42)					
Wholesaler	12.50	16.80	25.60	0.27	3.68
Retailer	0.83	1.04	27.95	0.12	3.15
Market III (3.11)					
Wholesaler	8.10	10.28	21.21	0.38	4.02
Retailer	0.65	0.92	29.35	0.15	2.19

ranged from 2.04 to 4.87 (Table 1). It was observed that retailers performed better (4.87) in Market I and wholesalers performed better (4.02) in Market III than other selected markets. Among the markets studied, the wholesalers in Market I faced

lot of difficulties in performing the fish marketing with very low efficiency index (2.04). This is mainly due to influence of agents who usually determine the fish price at the market.

Incidence of Extended-Spectrum Beta-Lactamase (ESBL) producing multidrug resistant *Escherichia coli* in seafood

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Escherichia coli is the most common commensal bacteria and they become opportunistic and obligate pathogens when coexisting with pathogenic strains (Martinez, 2009). The occurrence of E. coli in seafood is directly related with either faecal contamination or contamination by the infected handlers. Now-a-days, antibiotics are most commonly used in human therapies, farm animals and aquaculture (Cheong et al., 2014) which increase the incidence of resistant bacterial strains (Sapkota et al., 2008). Beta-lactam antibiotics are the most preferred treatment regimens for many of the infectious diseases (Bush and Jacoby, 2010). The widespread and indiscriminate use of antibiotics coupled with the transmissibility of resistance lead to the emergence of multidrug resistant (MDR) organisms like MDR-Extended-Spectrum Beta-Lactamase (ESBL) producing E. coli and become difficult to treat (Kang et al., 2005). In addition, E. coli may harbour the ESBL enzymes that confer resistance to most beta-lactam antibiotics mainly of extended-spectrum cephalosporins. ESBLs become easily transferable to other Enterobacterial species when encountered with it (Bradford, 2001). E. coli strains producing ESBLs are becoming multidrug resistant (MDR) and are considered to be one of the emerging pathogen world-wide (Ozcakar et al., 2011). ESBLs are a

group of enzymes and are encoded by genes on plasmids that hydrolyze all groups of beta-lactam antibiotics, including new generation group of cephalosporins mainly cefotaxime and cefriaxone (Perez et al., 2007). Whereas in the recent years, several studies demonstrated the prevalence of ESBL producing Enterobacteriaceae in food products such as meat, chicken, raw milk and fish (Koo and Woo, 2011; Ryu et al., 2012 and Ojer-Usoz et al., 2013). ESBLs genes enter and disseminate through the food chain via. direct contact with humans and animals which could contribute to the spread of these strains (Egea et al., 2012). There is no data available about ESBLproducing bacteria in food and aquaculture products and food of animal origin in Gujarat. Keeping this in view, the present study was carried out during September, 2011 to March, 2015 to monitor the presence of MDR and ESBL producing E. coli in seafood in Veraval, Gujarat, India. A total of 82 seafood samples (34.45%) were positive for presumptive E. coli on Tergitol - 7 out of 238 samples namely fresh samples (pomfret, horse mackerel, Indian mackerel, tuna, ribbonfish, seerfish, croaker, ghol, dhoma, sardine, shark, ray fish), dried fishes, from Veraval fish market and frozen samples such as prawns, cuttle fishes, cephalopods and surimi collected from fish processing industries in Veraval region, Gujarat. The *E. coli* counts ranged from $10^{1 \text{ to } 2}$ x 10^{2} cfu/g. About 140 E. coli isolates from 28 samples were confirmed on EMB agar and IMViC test and API 20E bioMerieux. One isolate from each confirmed sample was screened for ESBL producer in comparison with antimicrobial sensitivity. Eleven isolates (39.20%) were multidrug resistant (Fig.1) and four E. coli isolates (14.29%) were ESBL producers with triple ESBL detection Ezy MIC Strip (Fig. 2). Antibiotics were screened for determining MDR to cephalosporins (ceftazidime, cefepime, cefuroxime, ceftizoxime, cefoperazone, cefotaxime, ceftriaxone and cefoxitin), betalactam and beta-lactam inhibitors (amoxyclav, ticarcillin/ clavulanic acid, piperacillin/ tazobactam and ampicillin/sulbactam ampicillin) monobactam (aztreonam) and cephems and betalactam inhibitors (ceftazidime/ clavulanicacid), carbapenems (imipenem and meropenem), fluoroquinolones (ofloxacin, ciprofloxacin, gatifloxacin and levofloxacin), sulphonamides (trimethoprim/ sulfamethoxazole) aminoglycosides (amikacin and gentamicin). All these ESBL producers and ESBL-Amp C positive were multidrug resistant ≥3 classes of antimicrobials except carbapenem and aminoglycosides. The higher rate of resistance was found with ampicillin (39.29%), trimethoprim/ sulfamethoxazole (32.14%), ciprofloxacin (28.57%) cefepime (17.86%) and cefuroxime (17.86%). Whereas, all the isolates were sensitive to carbapenems (imipenem and meropenem), betalactam and beta-lactam inhibitors (piperacillin/ tazobactam and ampicillin/sulbactam) and aminoglycosides (amikacin and gentamicin) and these antimicrobial profiles clearly indicate the presence of ESBL producers. The results of this





Fig. 1. Antibiogram of $\it E.~coli$ isolated from the seafood with 24 antimicrobial agents (HiMedia, Mumbai)



Fig. 2. Detection of ESBL producing *E. coli* with Triple ESBL detection Ezy MICTM Strip (MIX+/MIX) EM 079 (HiMedia, Mumbai)

study indicate the prevalence of ESBLs and multiple antibiotic resistance ESBL producing multidrug resistant *E. coli* in seafood in Gujarat. Their presence revealed that the seafood might be the possible reservoir, transferring to human and posing serious threat to seafood consumers. So, strict hygienic measures are required to reduce the ESBL producing *E. coli* contamination in seafood.

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Nanoparticle-based antifouling coating for cage netting

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Fouling is a major problem in cage aquaculture and is more severe in tropical waters where the rate of fouling is very high. In cage culture, fouling results in reduced water exchange, low oxygen level and accumulation of faeces in the cage affecting the fish health. There is wastage of money, time and man hours in cleaning the clogged netting, and periodical net changing is a laborious process causing additional stress to the culture organism. Besides, heavily fouled netting adds extra weight, causes deformation of the net resulting in net failure and escape of the cultured organism incurring huge economic loss.

The use of metal based antifouling paints though effective, their use in cage aquaculture is undesirable due to possible effects on the caged fish and on the environment. Copper and titanium dioxide are known biocides and are extensively employed in antibacterial applications. Recently ICAR-CIFT, Cochin initiated a study incorporating

nano-sized copper oxide and titanium for fouling control in polyamide (nylon) netting. On application of the nano biocides, the nylon netting samples inhibited fouling even after 90 days of exposure to estuarine waters.

Coatings of nano-sized copper oxide (CuO) alone and mixtures of nano-sized (APS 40 nm)

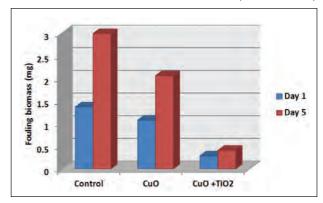


Fig. 1. Fouling biomass on nylon netting treated with nano biocides (0.005%)





Control CuO (0.01%) + TiO₂ (0.02%)

Fig. 2. Images of fouling accumulation on control and nano biocide coated nylon netting after 90 days exposure to estuarine waters

copper oxide and titanium oxide (TiO₂) in different concentrations were applied on nettings of polyamide (nylon) 210x2x2 having 30 mm mesh size. Netting, coated with mixture of copper oxide and titanium oxide showed better fouling resistance than copper oxide alone (Fig. 1).

Among different concentrations, netting coated with mixture of CuO (0.01%) and TiO_2 (0.02%) gave maximum fouling inhibition. Netting after 90 days exposure to estuarine waters showed very good fouling inhibition compared to control (Fig. 2).

The biocides play an important role in determining the attachment of microfouling and subsequent attachment of macrofoulers. The present study demonstrated that coating of netting with mixture of nano-sized CuO and TiO₂ was very effective in controlling fouling on nylon netting. The outcome of this study has a very positive effect on cage culture. The advantage of nano-sized particle is the requirement of less amount of particle and more coverage. Further studies with other netting materials such as high density polyethylene (HDPE) used extensively as cage netting is under progress.

Food loss from gillnets operated in Lakshadweep waters

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Depredation is the removal of fish (or part of the fish) or bait from fishing gear by predators (e.g. sharks, squids, cetaceans etc.), as distinct from predation, which refers to the capture of free-ranging fish (Anderson, 2014). Depredation on fish caught in fishing gear by predatory organisms is one of the reasons for food loss in fisheries sector. Quantifying depredation helps to estimate economic losses to industry, compare depredation levels in different gear and regions and helps to find out mitigation measures.

A study was carried out in Kavarathi island, Lashwadeep during January 2014 to monitor the food loss from gillnets operated for reef associated fishes. Kavaratti island of Lakshadweep, having an area of only 4.22 sq. km, is an atoll with unique biodiversity and specialized habitats. In the present study, depredation is considered as "the removal of gilled / entangled fish by crabs". Underwater observation for evidence of

depredation was carried out by operating a bottom set gillnet having 70 mm mesh size made of polyamide (nylon) monofilament yarn of 0.20 mm diameter. The net had a length of 200 m and depth of 5 m. Locally this fishing operation is known as *Balaidal* (gillnet set in the lagoon areas). Snorkelling, underwater observation by SCUBA divers and simultaneous recording with



Fig. 1. Underwater videography by study crew



Fig. 2. Fishes gilled in the gillnet

underwater camera (GoPro) and timed swims were used in our study (Fig. 1), which were effective in underwater observations. The fishing operation was carried out at a depth of 8 m and the net was soaked for 2 hours. Eight fishes (*Pomacentrus trilineatus* - 5 Nos., *Siganus corallinus* - 2 Nos. and *Halichorers hortulanus* - 1 No.) were gilled within 1 hour of setting the net (Fig. 2). After about 30 minutes, a crab (*Clappa hepatica*) approached the gilled fishes, and started feeding on them (Fig. 3). Within 2 h, 6 out of the 8 gilled

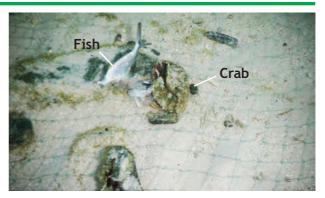


Fig. 3. Crab feeding on the fish caught in the net

fishes were partially depredated by the crab. The observations indicated depredation on fishes caught in gillnets by crabs. The depredation behaviour and habitat of crab in relation to the fishing gear need further investigations.

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Occurrence of Beaked sea snake (Enhydrina schistosa) in inshore waters of Cochin

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During demersal trawling operations onboard CIFT research vessels *M.F.V. Sagar Shakthi* and *M.F.B. Matsyakumari - II* frequent occurrence of sea snake (*Enydrina schistosa*) was observed from the inshore waters of Cochin during 2014-15.

Sea snakes are carnivorous in nature and their diet mainly consists of different types of fish, crustaceans, mollusks and eggs of various sea creatures. Venom of sea snakes is very strong and is used to kill their prey and to protect themselves in case of danger. Other than venom, some sea snakes produce enzyme that induces digestion of the prey from the moment of bite. Persistent myths about sea snakes include the mistaken idea that they can't bite very effectively. The truth is that their short fangs (2.5-4.5 mm) are adequate

to penetrate the skin, and they can open their small mouths wide enough to bite a table top. It is said that even a small snake can bite a human. Sea snakes can swallow a fish which is more than twice the diameter of its neck. Only a small proportion of bites are fatal to man, as the snake can control the amount of envenomation. Intense pain is not obvious at the site of the sea snake bite; 30 minutes after the bite there is stiffness, muscle aches and spasm of the jaw followed by moderate to severe pain in the affected limb. Then follows progressive CNS symptoms of blurred vision, drowsiness and finally respiratory paralysis. Sea snakes are poisonous but they are not aggressive creatures. Attacks of sea snakes are usually result of self-defense. Sea snake bites

Scientific classification

: Animalia Kingdom Phylum : Chordata Class : Reptilia Order : Squamata Suborder : Serpentes **Family** : Hydrophiidae Genus : Enhydrina **Species** : schistosa

occur on trawlers, when the snakes are sometimes hauled in with the catch.

The sea snake *Enhydrina schistosa* commonly known as Beaked sea snake is named due to the distinctive downturned, beak-like projection on the snout, at the front of the upper jaw (Fig.1). The scientific classification of the species is given in Table above.

Like other sea snakes, this species is highly adapted to life at sea, possessing a flattened, paddle-like tail for swimming, as well as valved nostrils, which can be closed when the snake is underwater. Sea snakes also lack the expanded belly scales that most other snakes use for moving on land. The body of the beaked sea snake is quite stout and vertically flattened, with a relatively small head. Adult is dull olive-green or greenishgrey on the upper side and whitish on the lower side, with dark crossbands that tend to fuse together towards the tail. The crossbands are widest on the upperside, tapering to points on the flanks, and usually disappear in older adults, which are of more uniform bluish-grey colour. They



Fig. 1. Sea Snake (*Enhydrina schistosa*) caught during trawling operation

average about 1.2 metres in length and have the deadliest venoms of all snakes. Beaked sea snakes give birth to up to 30 young ones each time they breed, but their death rate is so high that only a small proportion of the young ones survive to become adults. Despite their venom, these snakes are eaten by inshore predators, such as fish and estuarine crocodiles. These snakes are generally found in the coast and coastal islands of India. They are amongst the most common of the 20 kinds of sea snakes found in the region. They are active both during the day and at night. They are able to dive up to 100 m and stay underwater for a maximum of five hours before resurfacing. Sea snakes are equipped with glands to eliminate excess salt.

Sea snakes are usually caught in the depth range of 8-20 meters in live conditions. However reports say that duration of towe and quantity of catch had significant effect on within the trawl mortality. The crew alerts about the occurrence of snake in the net, even before the codend is lifted onboard. Once the codend is opened, the catch is released onboard, and the snake is lifted and released back by holding the tail and thrown overboard (Fig.2).



Fig.2. Lifting the snake for releasing

Foldable trap to exploit fishery in backwaters

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Fishermen of Andhra Pradesh State are using stake nets for exploitation of fishery in the intertidal regions. It is one of the oldest used gear in backwaters. These nets are fabricated with very small mesh and exploit maximum number of juvenile fish. The codend mesh size of this net is 5-8 mm. Fishermen have to spend more time for fabrication of nets and to reach stakes to tie the nets. ICAR-CIFT Visakhapatnam Research Center has developed a foldable trap to exploit fishery resources in backwaters (Fig. 1 and 2). In this design, four rectangular shaped plastic coated galvanized-iron (GI) square mesh of 25 mm mesh size are fixed to a frame made of 6 mm Gl rod. These four rectangular pieces are fixed with clamps and are arranged in a box shape and tied with HDPE twine. The length x width x height of the trap is 1 x 0.5 x 0.4 m. An iron mesh piece of 0.5×0.4 m is carved into funnel shape and tied in front as entrance. Other end of the frame having 0.5 m length and fitted with 25 mm mesh HDPE webbing is attached and closed as codend. When put to operation, shellfish and finfish were found entering through the funnel shaped mouth into the trap. Juveniles escaped through 25 mm square mesh of the trap and the adult fish remained in the last part of the trap.

The traps are foldable, easy to fabricate, easy to operate and durable. To maintain the ecosystem biodiversity and to conserve small juvenile fish and prawn, these foldable iron mesh traps are very much useful to exploit the fishery in backwaters and it serves as a substitute to the traditional stake nets.



Fig. 1. View of foldable trap before assembling



Fig. 2. View of foldable trap after assembling

Modified Gargoor trap for marine fishes off Saurashtra coast - Preliminary trials

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Trapping is a selective fishing technique with low impacts on the habitat. Fish trap is the

predominant gear used for the exploitation of demersal reef fishes. Traps are passive fishing

gears with enclosures to which fishes are lured or guided. Once guided inside the trap, the escape is made difficult by means of labyrinths or retarding devices like funnels or constructions. The cost of making simple traps is usually low and in many cases, cheap local materials can be used for its construction. The expenditure involved in setting and hauling of traps are usually not more, simple traps can be set and hauled even from a small canoe. In general, trapping and potting is good for the environment as it does little damage to the underwater ecosystem and it allows some control on unwanted bycatch. The most common types of finfish traps used throughout the world are Caribbean traps (arrowhead, "Z", "S", etc.), Round traps, Rectangular traps, "D"-shaped traps, collapsible traps, pelagic fish traps, North Atlantic cod pots and plastic multipurpose traps. Diverse types of traps like box traps, filter traps, tubular traps, plunge baskets, pound traps, etc. are in operation in the inland water bodies of India.

Enclosed transportable traps are by far the commonest type of traps used throughout the world. Although there are many types and shapes, including rectangular, circular, hexagonal, conical, semi-cylindrical, chevron-shaped and heart-shaped (arrowhead), the way they work is the same: fish enter the trap by one or several entrances or funnels and are prevented from escaping. Common parts of traps are frame, covering, funnels, door, bait holder, escape gaps and ballast.

Selection of fishing grounds

Selection of the type of trap and location for operation depend on the targeted species, their niche and behaviour. Most traps used in the tropics have been designed for fishing in reefs, rocky areas and on the rough bottom. So in the present study we also followed the same criteria. Saurashtra coast has many rocky patches not reachable to the conventional fishing systems like trawls, gillnets etc. Depths of these sites are normally not adequate for the operation of conventional fishing gears. For the present study, we have selected fishing area off Veraval coast, near Somnath temple, Gujarat, where the marine cage farming of ICAR-CMFRI is under progress. Before deploying the trap, hook and line fishing was done in the selected area to confirm the presence of demersal finfishes. Discussions were made with the local handline fishermen of Veraval. This discussion lead to the fabrication of five finfish traps used for the present study.

Construction and structural details of the experimental trap

Modified Gargoor fish traps were selected for the experimental trails. Galvanised iron was used for the construction of frame and HDPE webbing of 1.5 mm diameter was used for the body. Traps with single entrance and double entrance were made *viz.*, 40 mm mesh size (single entrance), 50 mm (single and double entrance), 80 mm (double entrance) and 100 mm (single entrance). Different stages of construction of trap are depicted in Figure 1. Diagrammatic representation of a typical trap is shown in Figure 2.



Fig.1. Various stage of fabrication of trap

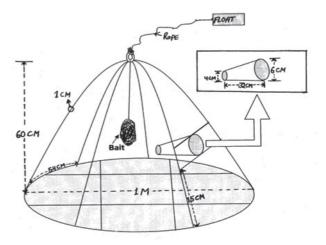


Fig. 2. Digrammatic representation of a typical trap with single entrance

Fishing trials

The experimental trials were carried out with the participation of *Siddi* tribes of Veraval. Ribbon-

fish, golden anchovy, cuttlefish etc. were used as wet baits. The bait wrapped in small meshed HDPE webbing was hanged in the centre of the trap, approximately 30 cm from the bottom panel of the trap. The traps were deployed at depths ranging from 5 to 10 m. Trap deployment at the study site is represented in Figure 3 and 4.

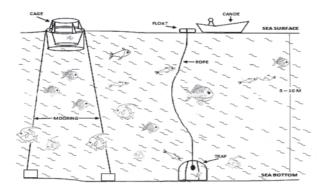


Fig. 3. Diagrammatic representation of study site and deployment of trap



Fig. 4. Deploying traps at the fishing ground

After socking for 24-48 hours, traps were lifted manually and the trapped fishes and crustaceans were collected, identified to species level and length-weight data recorded. A total of 17 trials were done starting from January to March, 2015. The sea water during the experimental period was comparatively clear.

Catch composition

In tropical areas, shallow-water reef and estuarine species are commonly caught with traps. Snappers, emperors, groupers, parrotfish, surgeon fish, squirrelfish, angelfish, tropical rock lobsters etc. were the common species caught by the traps. Catch details are given in Figure 5. Catch comprised of table sized commercially important demersal

fishes and crabs of size ranging from 150-400g belonging to the family Lethrinidae, Serranidae, Percidae, Portunidae, Actinopterygii, Xanthidae, Menippidae etc. Presence of the cage, left-over food material and rocky substratum might have helped in aggregation of the fishes to this site.

Among traps of different mesh sizes tried, better catch was in trap fabricated with 40 mm

Fig. 5. Major species caught in experimental fish trap



Bashful crab (Atergatis integerrimus)



Swimming crab (Charybdis lucifera)



Stone crab (Menippe rumphii)



Malabar grouper (Epinephelus malabaricus)



Brown sweet lip (Plectorhinchus gibbosus)



Emperor fish (Lethrinus sp.)



Crescent Perch bass/Tiger bass (Terapon jarbua)

mesh size followed by 50 mm (Fig. 6). In traps with other mesh sizes, catch was nil. Traps with

big mesh sizes facilitated entry and exit of small fishes which resulted in complete consumption of the bait.

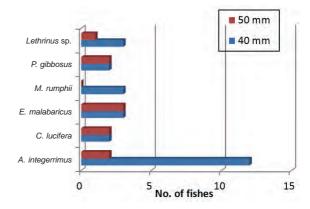


Fig. 6. Comparative catch in experimental traps of different mesh sizes

Conclusion

The results of the preliminary trials suggest that there is good potential of trap fishery along Saurashtra coast. Further research needs to be focused on aspects such as underwater observations on behavioural response of fishes to the trap, development of suitable material (low weight) for construction of trap, modification in trap entrance and bait preference.



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