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Contents

From the Editorial Board.....

The January-June 2018 issue of Fish Tech Reporter brings to you the recent research developments carried out at ICAR-CIFT on harvest and post harvest fisheries covered under 19 articles. In this issue, we are proudly introducing our breakthrough research findings: The rapid detection test kits for ammonia and formaldehyde contamination in fresh fish which can be definitely used as an on-site detection tool to control the use of hazardous chemicals in the domestic seafood sector. ICAR-CIFT is always instrumental in developing improved gear designs for the benefit of fishermen. The low drag shrimp trawl which helps in reducing the fuel consumption while trawling is a recent addition to those efforts. The article describing the use of biofouling resistant nettings may be of advantage to the cage fish farmers for reducing the cage maintenance cost. In an endeavor to promote responsible fishing, reports on the success story of square mesh codend adoption in the Sindhudurg coast of Maharashtra, introduction of off-bottom trawls in Goa coast and recycling of abandoned fishing nets by fishermen of Surashtra coast are featured in this issue.

ICAR-CIFT is progressively focusing on the use of safe and natural additives in fish products. Research notes on antioxidant packaging film with rosemary oil, marine astaxanthin as potential antioxidant and synthesis of gold nanoparticles using natural ingredients of aquatic origin are included in this issue. The current issue also features recent advancements in fish preservation and value addition including microwave vacuum drying and infrared drying of fish, fish-prawn sausage and dietary fiber -incorporated sausage, health beverage enriched with fish protein hydrolysates and ready to cook squid soup capsules. We introduce 'CIFTFISHPRO: An information system on ICAR-CIFT value added fish products' in this issue for the interest of seafood entrepreneurs.

We also report the presence of harmful methicillin resistant *Staphylococcus aureus* (MRSA) in seafood and immobilization of beneficial bacteria like sulphur oxidizing bacteria (SOB) for bio-remediation and collagenase producing *Bacillus* for fish waste utilization. We are sure that we are disseminating the latest innovations and interventions in the harvest and post harvest sector.

Low drag trawls for fuel saving

Sayana K.A., Remesan M.P. and Leela Edwin

ICAR-Central Institute of Fisheries Technology, Cochin

Fishing consumes 15 to 20 times more energy than it produces (Endal, 1980) and the average fuel consumption by the fishing industry is estimated at 15-21.5xl0^6 t (Thomson, 1988). Increased use of fuel intensifies the carbon foot print and green house gas effect which leads to global warming, climate change, etc. Fuel consumption assumes prime importance to fishermen due to hike in operational costs apart from its environmental effects. According to Tyedemers *et al.* (2005), world fishery fuel consumption is 50 billion (5 x 10^9) liters. There is an 8% increase in the contribution of fuel cost to the total operating expenses within a period of two years (Anonymous, 2011). Annual fuel consumption of mechanized and motorized fishing sector of India is estimated to be 1220 million liters (Boopendranath, 2000) and about 60-80% of the operational cost is contributed by the cost of fuel consumed.

Trawling is the most energy-intensive fishing activity and trawlers are one among the most fuel consuming fishing systems. Compared to passive fishing methods like gillnetting and long lining, trawling consumes five times more fuel and it is 11 times more compared to purse seining. To catch one kilogram of fish, trawling requires 0.8 kg of fuel while gillnetting requires 0.15, long lining 0.25 and purse seining 0.07 kg (Gulbrandsen, 1986). The fuel consumption of trawlers which depends on installed engine horse power and duration of voyage constitute 45 to 75% of operational expenditure. The resistance offered by the gear has a high effect upon speed of vessel and fuel consumption.

Under the National Agricultural Science Funded (NASF) project on Green Fishing Systems for Tropical Seas (GFSTS), ICAR-Central Institute of Fisheries Technology (ICAR-CIFT) designed and fabricated low drag trawls for fish/shrimp of head rope length 24.47 m 33.0 m, respectively. The drag reduction measures included in the design are increased mesh size and new material (Fig. 2). The material used is ultra high molecular weight polyethylene (UHMWPE). As UHMWPE provides same strength at a lower diameter, the twine size was reduced which results in reduced twine area. For evaluation of new designs, trawl nets using conventional material, high density polyethylene (HDPE) is also fabricated and used as control. The experiments for evaluating the new design were conducted onboard M.V. Matsyakumari II.

Fig. 1. Warp tension meter in use

From the trials conducted, the average reduction in drag of new design is estimated to be 17%. The drag of control and experimental gears at different operational parameters was also analyzed and UHMWPE trawls showed lesser drag than HDPE trawls (Fig. 3). The average fuel consumption per hour of trawling for HDPE trawls is estimated to be 30 liters and for UHMWPE trawls 26 liters (Fig. 4). The average reduction in fuel consumption was found to be 10%. The fuel consumption per kilogram of fish captured was also estimated to be 2.9 liters for HDPE trawls and 1.9 liters for UHMWPE trawls with an average

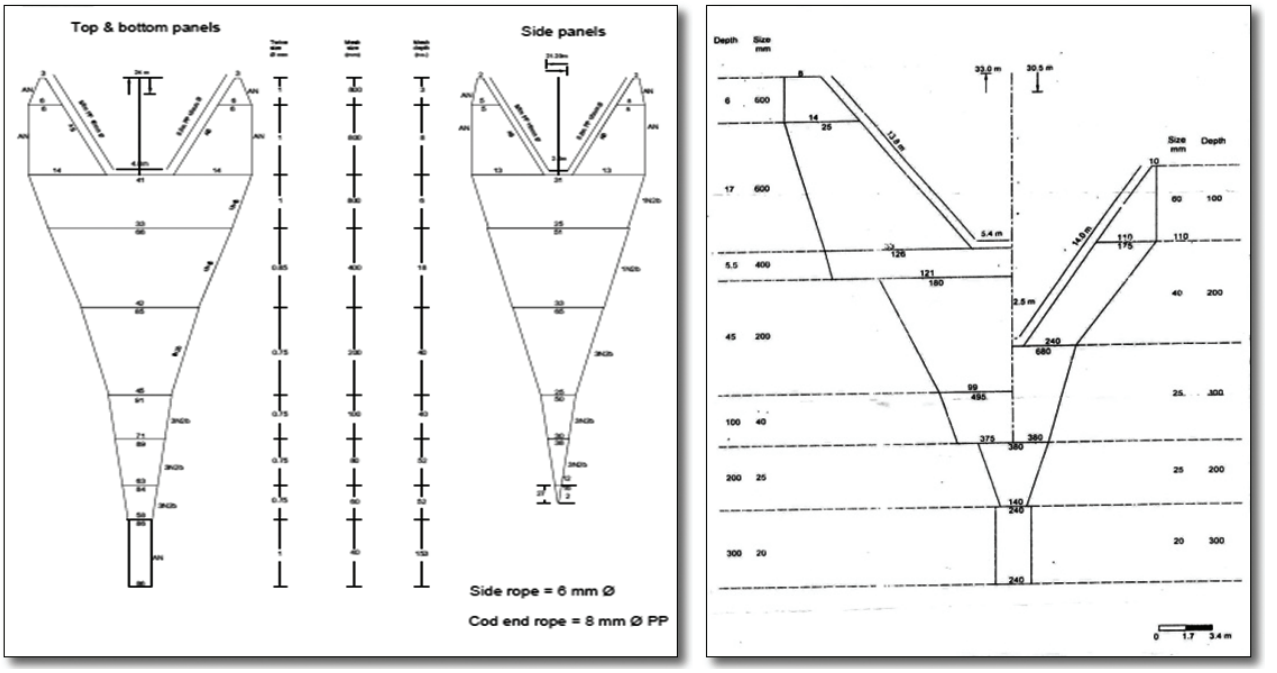

Fig. 2. Design of low drag trawls (24.47 m fish trawl and 33.0 m shrimp trawl)

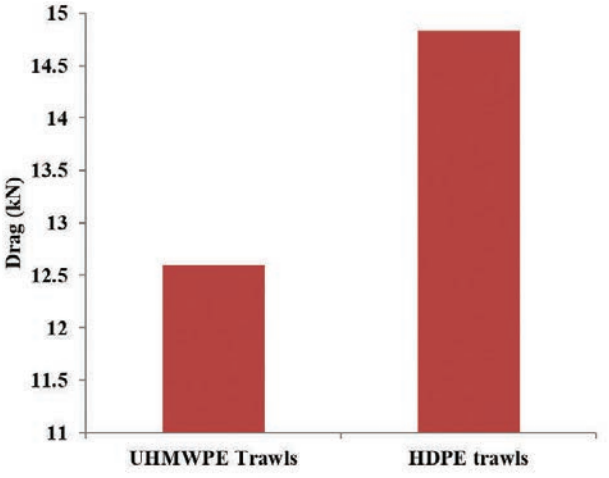

Fig. 3. Average drag of HDPE and UHMWPE trawls during one hour of trawling

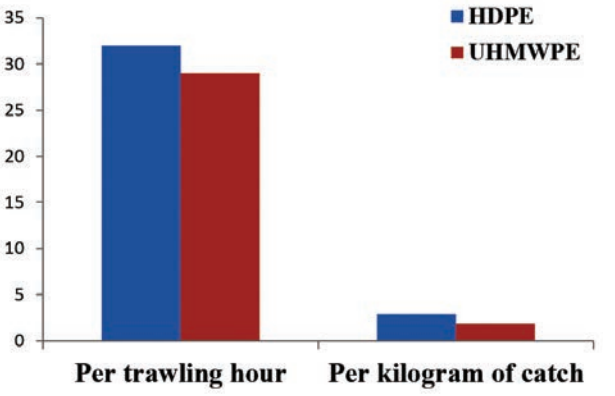

Fig. 4. Comparison of fuel consumption of conventional and low drag trawls

reduction of 35%.

As the name indicates, the drag and the fuel consumption of low drag trawls are 17% and 10% lower when compared to conventional HDPE trawls. Hence it is evident from the study that increased mesh size, reduced twine size and usage of energy saving material like UHMWPE will reduce the drag and thereby fuel consumption of trawlers considerably.

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Demonstration and operational efficiency of Off-Bottom Trawl System (OBTS): A new initiative in Goa by ICAR-CIFT

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Fishing contributes significantly to the economy of Goa, where bottom trawling is widely practiced. Several reports on the adverse effects caused by demersal trawling is available (Smith *et al*., 2000). The physical damage often caused by the gear on the sea bottom are highly detrimental to the bio-flora and fauna and it may take long time for recovery (Bijukumar and Deepthi, 2007; Gibinkumar *et al.,* 2012 and Bhagirathan *et al.,* 2014). Issues of bycatch in bottom trawling is also a major concern, especially in tropical countries like India due to the multispecies fishery (Velip and Rivonker, 2015). In this connection, ICAR-CIFT developed Off-Bottom Trawl System (OBTS), an ecofriendly trawl (Boopendranath *et al.,* 2011) capable of harvest of large demersal and semi-pelagic resources selectively with the aim to reduce bottom contact was demonstrated at Goa.

Experimental fishing trials of OBTS were conducted onboard a commercial trawler (S.F.X. Divine, L_{OA}-12 m, 120 hp) off-Chapora (Latitude 15° 36' and Longitude 73° 43') coast (Fig.1). Chapora is a major fishing ground for flat fishes and penaeid shrimps along the Goa coast. A 22 m, four seam OBTS having codend mesh size of 35 mm fitted with 65 kg suberkrub otter boards was used. Trawl operations were carried out in presence of trawl fishermen on 13 and 14 November 2016 along the coastal waters off-Chapora, Goa (Fig. 2). Five hauls of one hour duration each were conducted at depths ranging between 10 to 12 m. The haul-wise catch was quantified onboard and length and weight of individual species was recorded. Fourty one species of finfish and shellfish were identified in the catch of which 20 species belonged to demersal, 17 were pelagic species and two species of Molluscs and Crustaceans. Quantitatively, demersal, pelagic crustacean and Molluscan groups contributed 63.3%, 23.4%, 4.6% and 2.7%, respectively to the total catch. The count of commercial value (targeted) and low value bycatch (non-targeted) low value species belonging to pelagic fishes were 10 and 7, respectively. Similarly, for the demersal fishes it was recorded 10 species of each category. In the case of Crustacean and Molluscs, all the four species caught in the net were commercially important. The targeted commercial catch recorded for the OBTS was 70.3% and non-targeted low value catch was 29.7% of the total catch. There were no bottom dwelling organisms like shrimp, squilla and gastropods in the catch. Razorbelly scad (*Alepes kleinii*) formed the largest portion (19.1%) of the fish catch, followed by the yellowtail scad (*Atule mate*) and shrimp scad (*Alepes djedaba*) - 15.1% and 13.3%, respectively (Fig. 3).

The commercially important species like *Rastrelliger kanagurta* (1.5%), *Pampus argenteus* (2.2%), *Trichiurus lepturus* (2.2%),

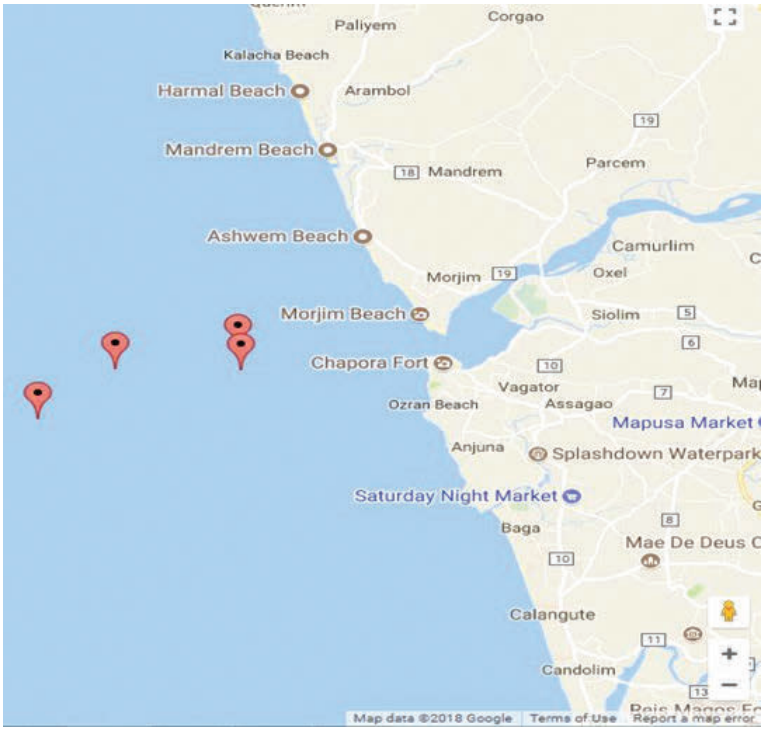

Fig. 1. Experimental trawling area off-Chapora

Fig. 2. Demonstration of rigging of OBTS to the fishermen off-Chapora

Fig. 3. OBTS catches on the deck

Lactarius lactarius (1.3%), *Epinephelus diacanthus* (3.7%), *Megalaspis cordyla* (3.8%), *Selar crumenophthalmus* (1.2%) and *Uroteuthis duvauceli* (1.4%), were comparatively less in the total catch. The average CPUE was 15.5 ± 6.9 (S.E.) kg/h. Average length of 13 commercially important species were analyzed for Minimum Legal Size and out of which 10 species were found to have average length well above the MLS proposed by Mohamed *et al.*, 2014 (Table. 1).

The OBTS performed satisfactory in terms of reducing non-targeted groups and Benthos. Switching to OBTS from bottom trawling during the non-shrimp season would help in harvesting the off-bottom resources without affecting the Benthic ecosystem. The inherent design characteristics of OBTS helps in reducing fuel consumption per kg of fish caught (Boopendranath *et al.*, 2011). This would improve the profits and also help in reducing CO_2 emission. More demonstrations in all maritime states and mass awareness programmes on OBTS through NGOs and fisheries co-operative societies are required for popularization of the technology.

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Table. 1. Comparison of mean size of commercially important catch from OBTS with MLS

Sl. No	Species	Mean size (cm)	MLS (cm) Mohamed *et al* (2014)
1	*Sardinella longiceps*	11.86	10
2	*Rastrelliger kanagurta*	16.89	14
3	*Pampus argenteus*	21.70	13
4	*Trichiurus lepturus*	47.84	46
5	*Lactarius lactarius*	10.82	10
6	*Epinephelus diacanthus*	13.21*	18
7	*Megalaspis cordyla*	9.00*	19
8	*Selar crumenophthalmus*	17.10	16
9	*Otolithes ruber*	24.60	17
10	*Otolithes cuvieri*	12.40*	16
11	*Portunus pelagicus*	14.90	9
12	*Portunus sanguinolentus*	12.70	7
13	*Uroteuthis duvauceli*	14.08	8
*Below MLS			

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Biofouling resistant polyethylene cage aquaculture nettings using polyaniline and nano copper oxide

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In Indian fisheries scenario, cage aquaculture is a fast growing sector to meet the increasing demand of high value fishes. Biofouling is a major concern in cage aquaculture where the infrastructure is exposed to a diverse array of fouling organisms with significant production impacts. Biofouling in cage netting causing clogging of meshes leading to increase in weight and drag, reduction of volume, restriction of water exchange, anoxic condition, increased stress, retarded growth of fish all of which adversely impact fish health and thereby increase the mortality rates also (Lai *et al*., 1993; Hodson and Burke, 1994). It has been reported that fouling in aquaculture adds 25% of the total project budget only for maintenance (Braithwaite *et al*., 2007).

ICAR-CIFT interventions in prevention of biofouling in aquaculture cages

Aquaculture cages are fabricated primarily with high density polyethylene (PE) webbings whose non-polar nature makes incorporation of antifouling biocides difficult. The surface of PE needs to be modified to develop strategies against fouling. Hence, antifouling formula of polar/ conducting molecule over PE was synthesized in our laboratory. Polyaniline (PANI) is a well-known conducting polymer since it is easy to synthesise, cheaper, has excellent stability, ability to sense and good adhesion with organic films (Arenas *et al*., 2012; Ullah *et al*., 2013).

Nanotechnology to prevent of biofouling in aquaculture cages

Polyethylene nettings (PE) of 25 mm mesh size were treated with freshly purified aniline. The polyaniline coated nettings (PE-PANI) were immersed in 0.02% aqueous nano copper oxide for 24 h, and air dried. The treated nets were exposed in open sea and estuarine environment to study the biofouling resistance.

Multi-location trials on biofouling resistance in polyethylene nettings

a) Open sea: Field exposure studies were conducted at Visakhapatnam (Open sea). Two treated webbings with its control were exposed in the open sea for a period of six months. After the exposure, treated webbing with its control was retrieved. The results showed significant biofouling resistance between the treatment and control samples (Fig 1). The control net was attacked by foulers (almost 80%), mainly the barnacles. The results reveals that PANI-nano copper oxide coated polyethylene as an excellent composition for antifouling in open sea.

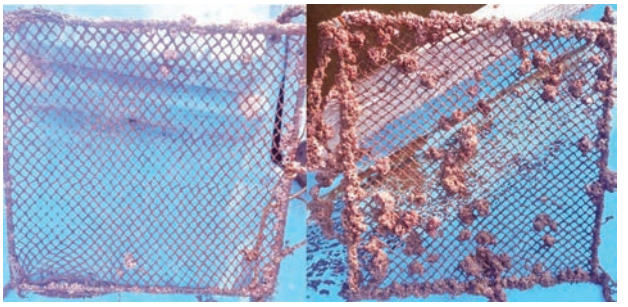

Fig. 1. Treated and control nets

b) Aquaculture farm: Field exposure studies on PANI-nano copper oxide coated polyethylene for antifouling was conducted at an aquaculture pond in Kuzhippally, Vypeen. A polyaniline copper oxide composite was synthesized *in situ* and coated over polyethylene webbings used for cage nets. The webbings with its control were exposed in the aquaculture ponds for a period of seven months.

Initially the algal fouling was significantly more in treated than in untreated sample. Polyaniline with ammonia may act as an initial source of nitrogen for the algal growth. But gradually in the succeding months algal fouling showed a declining trend in the treated sample. However control showed a steep rise in algal biomass (Fig.2 and 3). Same trend was observed in the seventh month also.

Conclusion

The study emphasized the importance of

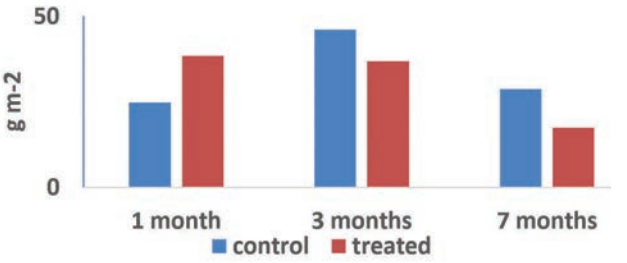

Fig. 2. Accumulation of biomass over the exposed aquaculture cage nets at Kuzhipally, Vypeen

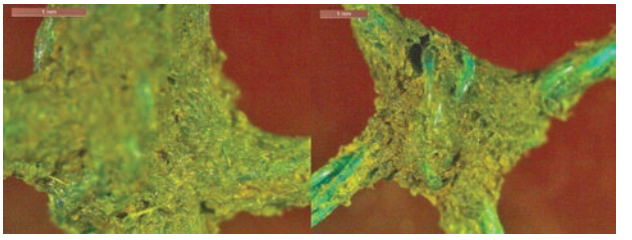

Fig. 3. Control and treated mesh after field exposure (16 X)

in situ synthesis of PANI over PE nettings (PE-PANI) and application of nano copper oxide as a biocide to combat marine biofouling. The nano copper oxide present in the matrix acted as a point source above the electron clouds of polyaniline, preventing initialization of biofilm. The results highlighted the potential application of polyaniline to modify the non-polar surface of polyethylene to load active biocides to prevent fouling in cage aquaculture. The results of the field exposure studies, revealed a great potential for polyaniline with nano copper oxides against biofouling in aquaculture cagenets in sea/ estuarine conditions. This technology has to be further promoted for commercialization.

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Adoption of square mesh codends for the trawl fishery: A success story along Sindhudurg coast, Maharashtra

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With over 24,000 tonnes of fish catch, the Sindhudurg coast of Maharashtra is an important fishing ground along the Indian coast. Trawlers (314 Nos. in total) operating from three landing centres *viz.*, Vengurla, Malvan and Devgad, contribute to the bulk of the landings from this region (CMFRI, 2012). The trend of increasing the length of the vessels with concurrent increase in the engine power is not reported in this region. The L_{OA} of the trawlers ranged between 12-15 m and are fitted with 104 HP marine diesel engines. Though, there are no reports of bycatch generated by trawlers at Sindhudurg, Pramod (2010) reported that nine lakh tonnes of discards is generated along Maharashtra coast, which is 12 percent of weight of the total landings. A project funded by United National Development Programme (UNDP), catering to all aspects of the economy of Sindhudurg District was taken up by the Government of Maharashtra and was implemented by the Mangrove Cell of the Forest Department. As part of this project ICAR-CIFT was asked to take up a project to address the problem of high bycatch incidence in trawlers in the region.

The study started with an initial survey covering all the trawl landing centres of the region to collect details regarding the specifications of the most common gears used and other operational parameters in the trawl fishery. It was observed that the trawl nets of this region are smaller in size when compared to the dimensions of trawl nets used along the west coast of India (Saly N. Thomas *et al.*, 2015). The mesh size of the webbing used in the codend ranged from 15 to 25 mm. The composition of bycatch consisted of 70-75 % of juveniles of ribbonfish, sciaenids and squids (Fig. 1), depending on the season of operation. The highest bycatch was noticed during January (56 kg/ haul), followed

Fig. 1. Juveniles of commercially important species in the bycatch

by December with a catch rate of 43 kg/haul.

Apart from the monsoon ban that is religiously followed, no other technical measure was adopted in the fishery. Apart from the demonstration of the Juvenile Fish Excluder Cum Shrimp Sorting Device (JFE-SSD), no other bycatch reduction device was ever tried in this region and there were no reports on the selection properties of trawl codends from this region.

As a prelude to implementing the project, 14 awareness programmes were conducted for the fishermen and the officials from the Department of Fisheries, in which the different responsible fishing techniques developed by ICAR-CIFT were explained using audio-visual aids. Feedback data using structured questionnaires was collected, from the trainees. Many fishers who attended the programme raised their apprehension regarding the escape of commercial catches from the square mesh codend. Based on the feedbacks and the interaction with the fishers, it was finally decided to demonstrate the operation of Square Mesh Codends (SMC) and Semi-Pelagic Trawling System (SPTS) on-board the trawlers of fishers.The trials were conducted on-board three commercial fishing vessels off Malvan, Vengurla and Devgad fishing harbours.

To make the fishers visually aware of the escapement, a small mesh cover was woven over the square mesh codend, to collect the escapees from the codend. The cover also helped to collect the length-frequency and finally to derive the selectivity parameters of some commercially important species. The catches from the codend cover were kept separately and analyzed for its composition to convince the fishers regarding the damage to ecosystem, due to the use of small mesh in the codend.

A total of 38 comparative hauls of 95 hours each, using trawl nets installed with square and diamond mesh codends of 35 mm mesh size were carried out. The average CPUE recorded in the square mesh codend was 19.48 $kg.h^{-1}$ and trawls using diamond mesh had an average CPUE of 18.77 $kg.h^{-1}$. The length-frequency of the species caught in both the codends were also analyzed to show the better selectivity of square mesh codends. The mean lengths of 12 out of the 15 commercial species studied, were higher by a minimum of 7.8% in the square mesh codend compared to the length of the same species caught in diamond mesh (Fig. 2). The results of the study, convinced the fishers that there is no significant revenue loss occuring if square mesh codends are used.

The escapement from the square mesh codends was between 2.5 kg per hour, which was about 3.5 percent of the total catch in the codend. A total of 83.6 kg of juveniles were released by the square mesh codends during the study. The price of the bycatch was ₹ 15 per kilogram, and the value of excluded catch was about ₹ 22.5 per haul.

Arathy Ashok and Madhu (2017) carried out a study to understand the level of adoption of responsible fishing operations along Sindhudurg coast and results showed that 100 percent of the fishers interviewed were aware of the technology and about 40 percent of the fishers had already adopted the new technology. Since the technology involves natural resource conservation in which benefits are often not perceived directly, it is often recommended to have initial support mechanisms for adoption. Taking this into consideration, the UNDP-GEF project, followed by the Government of Maharashtra under the District Development Fund had supplied one square mesh codend each for the 314 trawlers operating along the coast. All the trawlers have since then started using square mesh codends during their operations.

Regular training programmes for conversion of diamond mesh to square mesh codends were imparted to the net makers and fishers in which 347 fishers were trained in 27 programmes. Based on the outcome of the study, the Government of Maharashtra vide its notification dated the 10th January, 2017, has amended the Maharashtra Marine Fisheries Act, 1981, directing that no trawl gear having less than 40 mm square

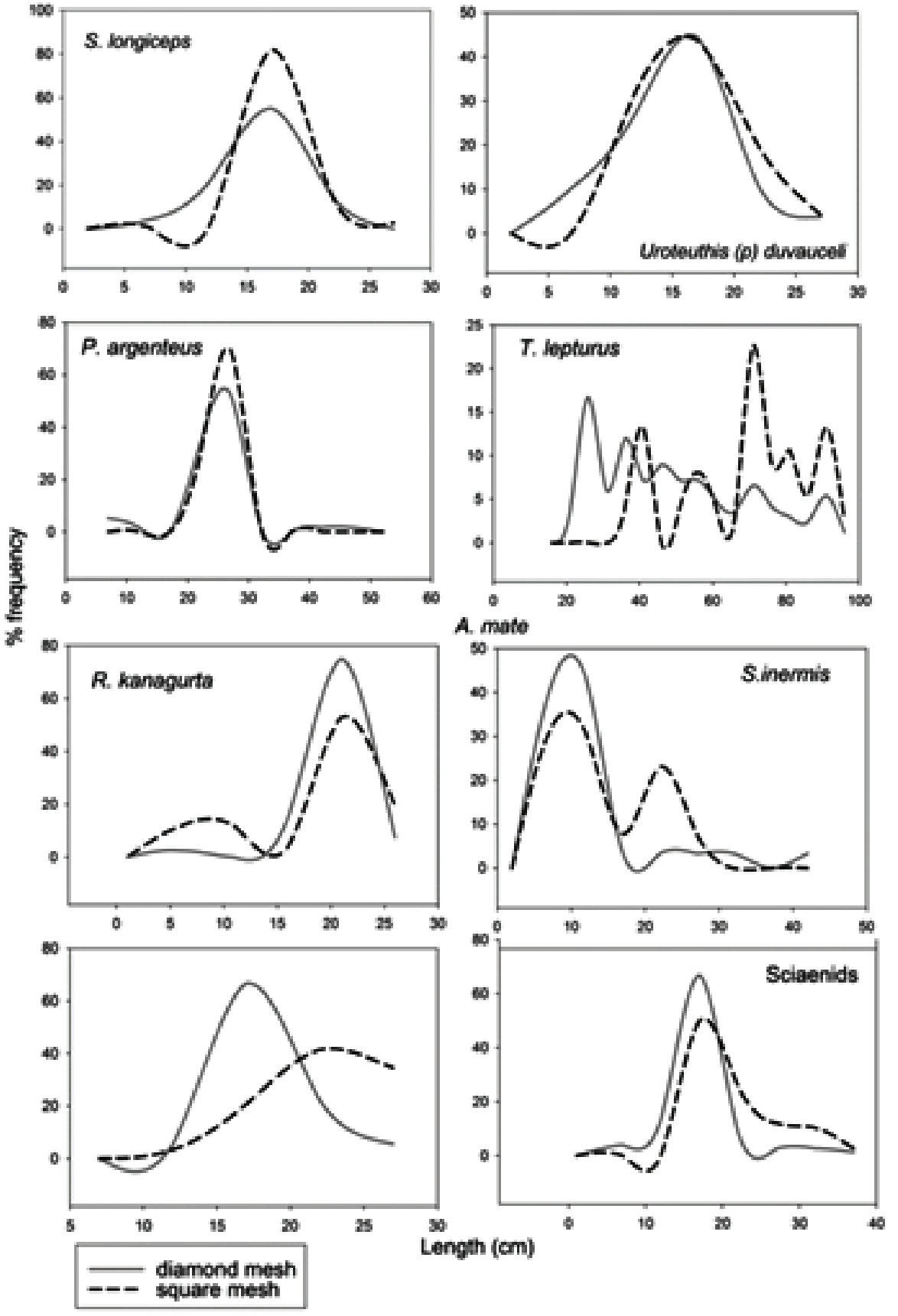

Fig. 2. Size frequency distribution of major species in the diamond and square mesh codends

mesh nets at codends shall be operated by any mechanized fishing vessel.The government of Maharashtra has decided to supply square mesh codends on a buy-back scheme to all the 5613 trawlers operating along Maharashtra coast.

The stipulated mesh size for the codend is 40 mm, which, when used during the shrimp season may lead to escapement of small quantities of shrimp. So optimization studies would be required to finalize the legal mesh size of codends for use during shrimp season.

This approach, for implementation of gear-based technical measure, was very successful, since it actively involved the Department of Fisheries, Department of Forests and support of the Fishermen Societies and can be taken as a model for implementation of Bycatch Reduction Devices elsewhere.

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Development of antioxidant packaging film using Rosemary Essentail Oil (REO) and chitosan

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Active packaging is a type of packaging which changes the condition of the packed food to extend shelf-life or to improve safety or sensory properties, while maintaining the quality of packaged food. Typically, this refers to the incorporation of certain additives into the packaging systems with the aim of maintaining or extending product quality and shelf-life. Packaging can be termed active when it performs some desired role in food preservation other than providing an inert barrier to external conditions. Most important active packaging concepts include O_2 and ethylene scavenging, CO_2-scavengers and -emitters, moisture regulators, antimicrobial packaging concepts, antioxidant release, release or absorption of flavours and odours etc. Recently, with increasing health concerns of consumers, current packaging research is mainly focusing on the use of natural compounds such as chitosan, essential oil etc. either as edible coating or active agents in packaging material to preserve and prolong the shelf life of food. Among the essential oils, rosemary oil has proven antioxidant as well as antimicrobial properties

against spoilage and pathogenic microorganisms. In fishery products oxidation is the major quality problem affecting its quality and shelf life due to high amount of poly-unsaturated fatty acid (PUFA). Though synthetic antioxidants are being used to prolong the shelf life of food products, it may lead to shorter life span to consumers due to their ill effects on health. This resulted in demand for natural antioxidants either to use directly or as packaging film. A study was undertaken to develop an antioxidant film incorporating rosemary essential oil in chitosan and its characteristics were evaluated. Chitosan with degree of deacetylation of 90.28% and viscosity of 246 cP was used to develop an antioxidant film incorporating 0, 0.5, 1 and 2% rosemary essential oil. Thickness of the film ranged between 24 – 27 µm. The surface morphology of the film studied using SEM revealed non-porous film with plain texture with even surface for chitosan film without rosemary essential oil (Fig. 1). Addition of rosemary essential oil resulted in uneven flaky surface which increased with increasing level of essential oil. Oxygen transmission rate, tensile strength and elongation at break decreased with increasing level of rosemary essential oil whereas solubility increased with increased essential oil level. The solubility of control chitosan film was 11.7% compared to 16.7 to 18.6% for rosemary extract added chitosan films. Addition of rosemary extract affected the colour values of the films. The lightness showed a decreasing trend with the increase in rosemary extract level in the films. The whiteness index showed a decreasing trend whereas yellowness index showed an increasing trend with the increase in rosemary

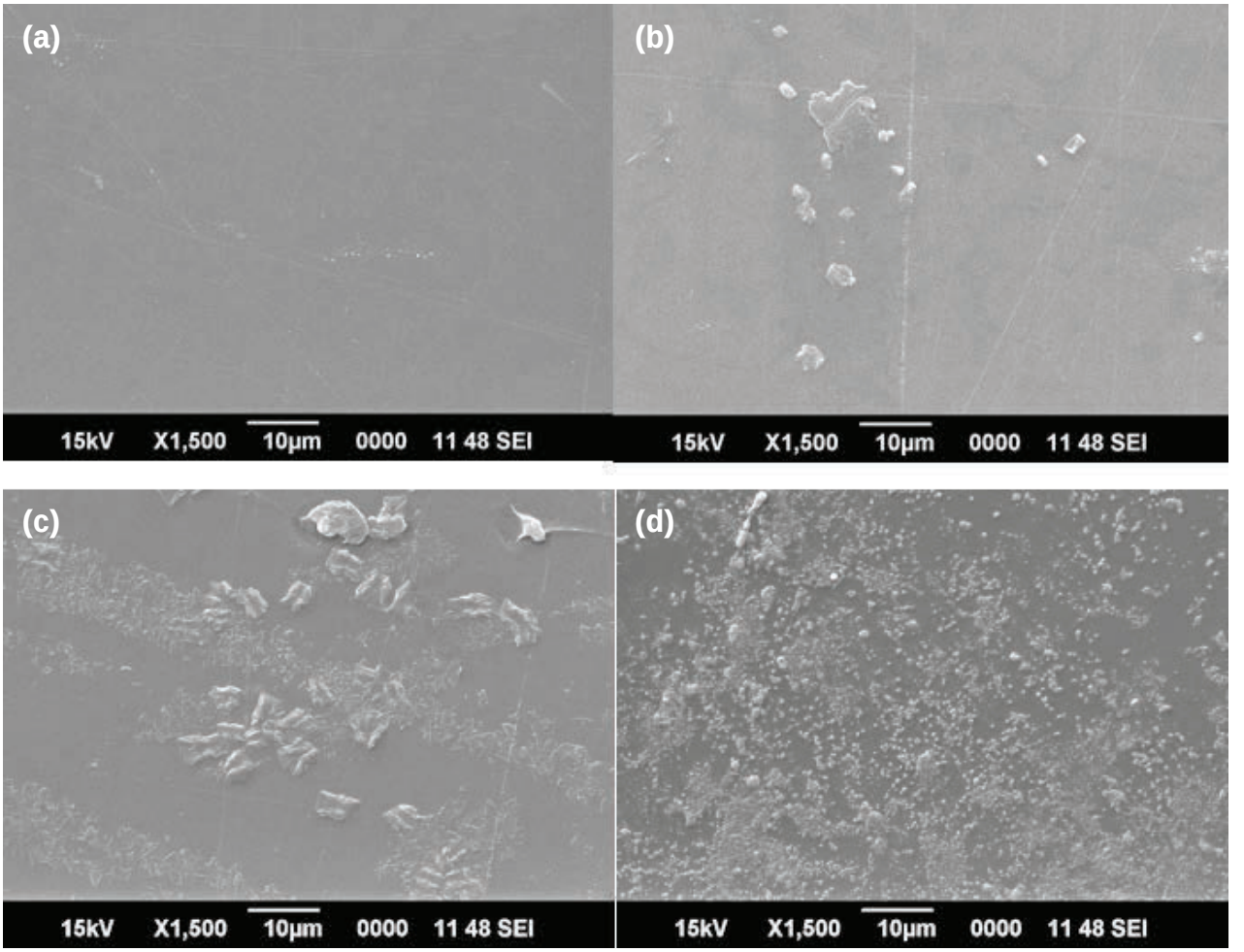

Fig 1. SEM images of surface morphology of chitosan films (a) chitosan film with 0.5% REO (b) 1% REO (c) and 2% REO (d)

extract levels in the films. Yellowness of the film increased significantly to 39.23 for 2% rosemary essential oil compared to 1.99 for control chitosan sample. Both DPPH activity and total phenolic contents increased with the increase in rosemary extracts level in the films (Fig. 2). Total phenolic contents of the film varied between 0.1 to 11.28 mg gallic acid per g film. DPPH content of control sample was 1.3%, whereas it varied between 12.42 to 22.51% in chitosan film with rosemary extract. The results demonstrate that rosemary incorporated chitosan films can be used for packing food products including fishes to enhance the oxidation stability.

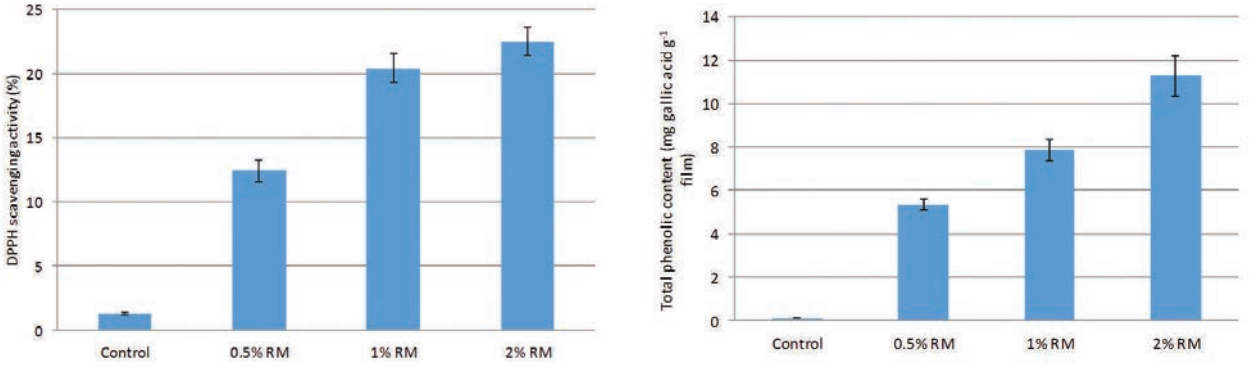

Fig 2. DPPH scavenging activity and total phenolic content of chitosan film incorporated with different levels of REO

Astaxanthin: A promisive antioxidant and UV protective agent

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Astaxanthin (3,30-dihydroxy-b,b-carotene-4,40-dione) is a naturally occurring carotenoid pigment belonging to class of phytochemicals, and is found in certain animals and plants. It is a powerful free radical scavenger and thereby naturally reduces the level of free radicals in the body. This uniqueness of astaxanthin may be effectively explored for its use as antioxidant and UV protective agent, where free radicals are primarily responsible for the deteriorative changes. This activity is mainly due to its unique molecular structure; polar ionic rings and non-polar conjugated carbon-carbon bonds. The main sources of astaxanthin are krill, algae, red trout, shrimp, crab and lobster. The intense colour of these species is on account of their richness in this red pigment.

Astaxanthin was extracted from shrimp head waste and characterized for antioxidant and UV protective properties. The highest yield was obtained with hexane 48.93 µg/g wet shell extract, followed by acetone, methanol, ethanol and chloroform. The extracted astaxanthin was further dispersed in virgin coconut oil (Fig. 1). The stereo-microscopic image of the extracted astaxanthin indicated spherical granular geometry (Fig. 2). The antioxidant activity assays indicated high DPPH free radical scavenging activity of 0.4 µg IC_{50}, Fe reducing activity (1.25 : A700/mg) and metal chelating activity of 34% at 70 µg/ ml.

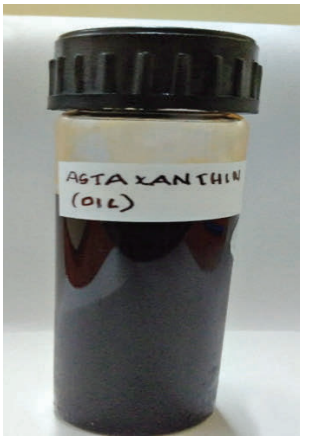

Fig. 1. Astaxanthin in virgin coconut oil

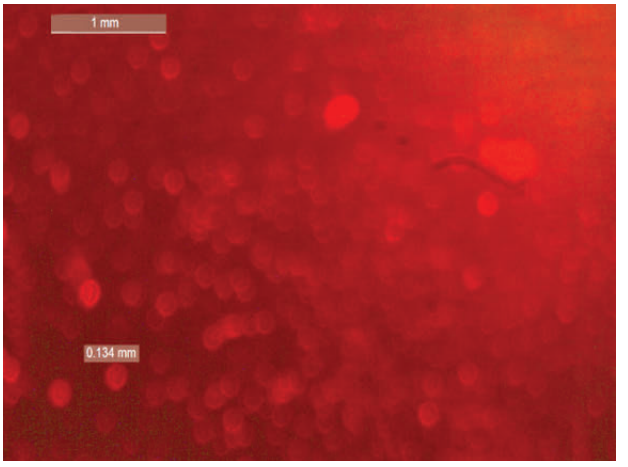

Fig. 2. Stereo-microscopic image of astaxanthin extracted from shrimp head waste

Thermal denaturation profile indicated a rapid rate of denaturation above 60 °C.

UV Spectra of astaxanthin indicated high absorption at UV range of 200-400 mm which suggests its potential to be used in as cosmetic formulations as UV protective agent. The UV protective effect of astaxanthin was evaluated on *Staphylococcus aureus* ATCC 25923. Results indicated good UV protective effect for the extracted astaxanthin in terms of cell viability. Bacteria grown in astaxanthin-incorporated culture media gave protection to the colonies even after exposure to UV radiation for 48 hours (Fig. 3). Further studies also indicated that the UV protective effect of astaxanthin was not altered during accelerated storage conditions.

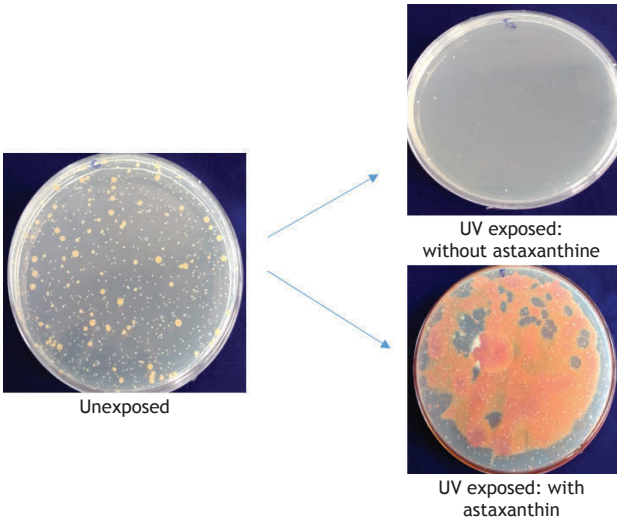

Fig. 3. UV protective activity of extracted astaxanthin on Staphylococcus aureus

Optimization of prawn pulp-incorporated fish sausage using mixture response surface methodology

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A study was undertaken to optimize the combinations of surimi and prawn pulp for the development of prawn pulp-incorporated fish sausage in order to enhance the utilization of small varieties of shrimps. Fish mince was taken from Thread fin bream and prawn pulp was taken from *Metapenous dobsoni* (Thelly chemmeen) for the development of combination sausage. A D-optimal mixture design for 10 different combinations of surimi and prawn pulp was

Table 1. Ingredient combination of sausage

Ingredients	% Weight
Surimi and Prawn pulp	86.7
Salt	2.15
Spice mix (mint: ginger: chilly)	0.9
Black pepper powder	0.4
Corn flour	7.8
Garlic	0.2
Cinnamon and clove powder(1:1)	0.3
Vegetable oil	1.5

formulated with different proportions of surimi and prawn pulp. The ingredients of combination fish sausage is given in Table and sausage was prepared by standard procedure.

Quality response variables *viz*: Texture profile analysis (TPA), Water holding capacity (WHC) and Colour values were measured for each combination by standard analytical procedures. Sensory score in terms of Overall acceptability (OA) was recorded in 9 point Hedonic scale. Linear, Quadratic and Cubic mixture response surface regression models were fitted to the quality response variables using OLS method and goodness of fit of the models were assessed by R2 values (Myers and Montgomery, 2002). The functional forms of Linear, Quadratic and Cubic models are given below:

Linear: $Y=\sum_{i=1}^{q}\beta_i x_i + e,\ i=1,2$,

Quadratic: $Y=\sum_{i=1}^{q}\beta_i x_i + \sum_{i<j=2}^{q}\sum \beta_{ij} x_i x_j + e,\ i=1,2$,

Cubic: $y=\sum_{i=1}^{q}\beta_i x_i + \sum_{i<j=2}^{q}\sum \beta_{ij} x_i x_j + \sum_{i<j=2}^{q}\sum \delta_{ij} x_i x_j (x_i - x_j) + e,\ i=1,2$

where Y is the response variables and are linear, quadratic and cubic regression coefficients and e is the error term. The above models were used to predict the response variables. Linear model was found to be best fitted model for springiness, quadratic model for hardness 1, L* and a*; and cubic model for WHC, b* and OA.

Fig. 1. Optimized prawn flavoured combination sausage

The desirability score was computed based on the predicted values of the response variables and the optimum combination was found to be 83.4% surimi and 3.3% prawn pulp; and the corresponding desirability score was 0.99. The combination sausage prepared at this optimum level was also found to have maximum consumer acceptability.

Reference

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Development of dietary fiber-incorporated fish sausage

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Sausage is a major item of new generation fast food, because of overwhelming young generation craving for its superb taste, fabricated texture, brilliant colour and high nutritional quality. Fish sausage prepared from the mixed mince of under-utilized fishes would be a healthy option for the new generation. The addition of dietary fiber to fishery products is of particular interest as a means of improving the functionality of food products and as a component of new functional foods. The addition of dietary fiber would help to further improve health features of consumer by reducing cholesterolemia, modifying the glycemic response, increasing nutrient availability etc. Therefore, a study was undertaken to develop fiber-enriched tuna sausages by optimizing different combinations of oats and wheat dietary fibers by considering the quality parameters.

The chilled yellowfin tuna (*Thunnus albacares*) blocks were purchased from fresh fish cutlet in Cochin, Kerala. Fish blocks were split into small pieces and washed immediately in potable water and removed all impurities and bones and made into mince by using meat bone separator. Two different types of dietary fibers were used for the preparation of dietary fiber-incorporated tuna sausage samples. The selected VITACEL® brand dietary fibers were oats fiber HF 401-30 and Wheat fiber WF 600R supplied by M/s Rettenmaier India Pvt. Ltd. Sausages were prepared according to the standard recipe by replacing only desired percentage of fish mince with oats and wheat dietary fibers. The common ingredients for the preparation of a standard sausage like salt (2.50%), sugar (1.50%), potassium sorbate (0.10%), polyphosphate (0.20%), guar gum (0.01%), spice mix (0.10%), spice concentrate (1.50%), fat (5%), corn starch (9%), ice (10%) were used for all these samples in same proportion. Recipe ingredients used in this preparation were of food grade quality.

An experimental design with 13 runs was formulated for the development of dietary fiber-incorporated tuna sausage by substituting tuna mince with fibers. Fish mince ranging from 60 to 70%, wheat and oats fiber ranging from 0 to 5% were used for the formulation of the experimental design with a constraint on sum of mince, wheat and oats fiber to 70%.

The dietary fiber-incorporated tuna sausage was developed by standard procedure. After mixing, all ingredients were stuffed into commercially available polyamide casings (35 mm diameter) to a length of 100 mm by using a hand operated sausage stuffer. Sensory analysis of the sausages was done by a five member expert panel using a 9 point Hedonic Scale prescribed. The score of 9 denoted the quality description ''likes extremely'' and 1denoted ''dislikes extremely''. A score of 4 was considered as the margin of acceptance. Sausages were cut into uniform pieces and warmed in a microwave oven for 1.5 min. and presented in coded plates. Panelists were asked to score on appearance, colour, odour, flavor, taste, texture and overall acceptability of the samples (Rahman *et al*. 2017).

The best combination was selected based on the sensory and physical parameter evaluation of 13 combinations. The sensory scores of 13 combinations are given in Fig. 1. It was found that sample Number 12 (S12) and sample Number 13 (S13) have highest sensory score compared to the others (Fig. 2). Apart from sensory scores, physical parameters of S12 and S13 were also found to be the best.

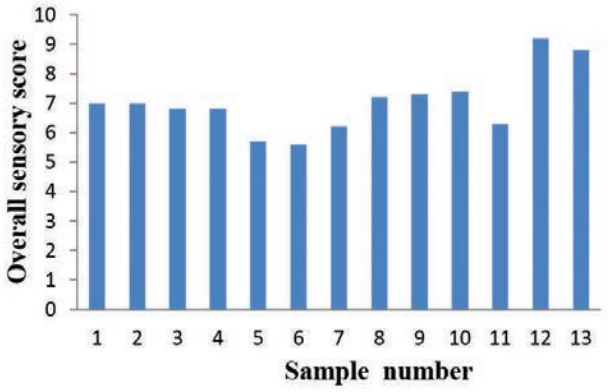

Fig. 1. Overall sensory score of sausage samples

The optimum combination of sausage was found to be 66.8% fish mince with 1.6% each of oats and wheat fibers (S12); and 62.5% fish mince with 2.5% wheat fiber and 5% oats fiber (S13). On the basis of the proximate evaluation of samples, control and Sample S12 had no significant change in the proximate values. The Sample S12 had 18% carbohydrate,15% protein, 7% fat, 1% ash and 59% moisture. But Sample S13 had higher amount of carbohydrate, protein and less moisture compared to control. The proximate composition of Sample S13 was 23% carbohydrate, 16% protein, 7% fat, 1% ash and 53% moisture.

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Control Sample S12 Sample S13

Fig. 2. Dietary fiber incorporated tuna sausage samples (S12 and S13) and control

Quality evaluation studies in ready-to-use squid soup tablets

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Development of value added products from seafood is an important need in fish processing industry. There is an increasing demand for ready to cook or ready to serve type seafood products.

Squids have been consumed by humans for centuries. Although squid is widely consumed, people still do not understand or under-estimate its nutritional value. It is found that squid may be one of the greatest untapped sources of protein in the marine environment. On comparing squid to other edible marine life, it is evident that squid has a larger ratio of edible parts to the whole body. In squid, the edible portion represents 60-80 percent of the weight of the animal, the body being 50 percent and the arms being 30 percent. Squid contains all eight essential amino acids in nearly proportioned quantities. Squid meat is having a characteristic sweet taste. Since the quantity of fat is too low, the amount of cholesterol present does not constitute a health hazard, even to those who are on a diet. Squid meat also has higher levels of zinc, manganese and copper than many other types of seafoods. The high nutritional quality of squid has given the idea of developing a dehydrated squid soup tablet which gives instant soup on mixing with boiling water.

Fresh squid procured from Cochin Fisheries Harbour were iced immediately in 1:1 ratio and brought to the laboratory in insulated boxes. The edible portions were cut into small pieces and were washed thoroughly in running tap water. The cut meat was dried at 50 °C and the dried meat was powdered. The other ingredients *viz.*, onion, garlic, ginger, pepper, cumin etc. were cleaned, dried and powdered. The vegetables like beans and carrots were chopped into fine pieces, blanched and dehydrated. The base flour used was maida and corn flour. Each ingredient of the required quantity was weighed out into a clean vessel and mixed well. Dough was made by adding water and butter and flattened into even layer from which soup tablets were cut out using a round mould. The tablets were dried at 60 °C to a moisture content of ≤10%. It is further cooled, packed in small LDPE pouches and stored at ambient temperature. The storage studies of tablets have shown that it has an extended shelf life of one year. Figure 1 and 2 depicts the dehydrated squid soup tablet before and after packing.

Fig. 1. Dehydrated squid soup tablets before packing

Fig. 2. Dehydrated squid soup tablets after packing

The prepared soup powder tablet was having 22% protein and 1.43% fat. It's keeping quality was assessed for a period of 12 months. The squid soup tablet was acceptable throughout the storage period of 12 months and was scored within the levels of acceptance during sensory evaluation. The limit of acceptability for sensory evaluation was fixed as 5. Figure 3 illustrates the changes in sensory attributes in soup tablet on

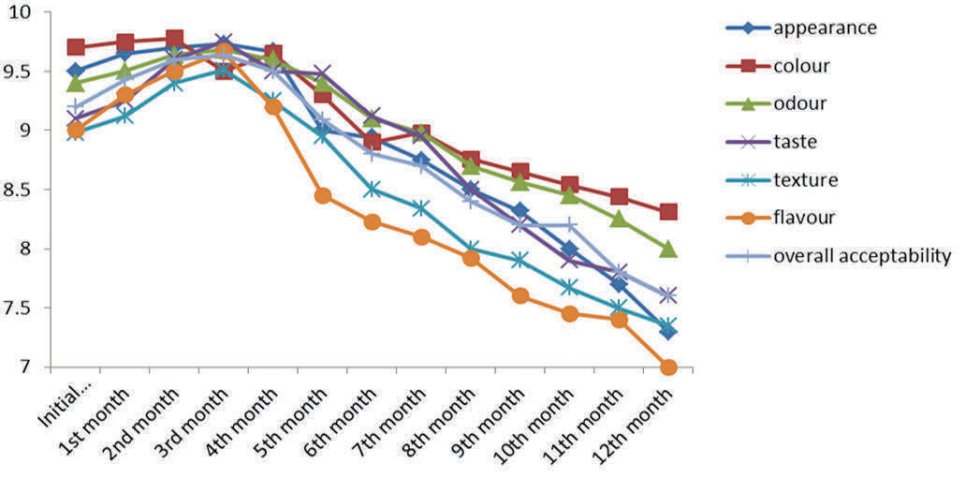

Fig. 3. Change in sensory attributes in soup tablet on storage at room temperature

storage at room temperature.

The chemical spoilage indices *viz*; TMA, TVB-N, TBA, pH, FFA, a_w were found to increase as the storage period advances. All these parameters were within the acceptable value till the end of storage period of 12 months. Total plate count (cfu/g) of the soup tablet increased during the storage period and it reached upto 6.8 X 10^5 cfu/g. There was only 3 log cycles increase during the storage period. Bacteria of public health significance were assessed during storage period. *E. coli, Vibrio* sp., *Salmonella* sp. and *S. aureus* could not be detected in the samples during the storage study.

Utilization of yellowfin tuna protein hydrolysate in health beverage formulation

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Seafood is an easily available and cheapest food source meeting the protein requirements of approximately 2.9 million people, globally. There is a high potential in marine processing industries to convert and utilize this food and their by-products as valuable functional ingredients. Among the by-products, hydrolysates or bioactive peptides can be utilized as a potential source of natural ingredient and in this context more focus is given by researchers on improving the bio-availability and bio-accessibility of these marine protein hydrolysates for validating as functional ingredients for healthy foods. Functional foods defined to be those with specific health benefits, hold a strong market position world-wide and the functional beverage sector accounts for approximately 12.5% of the world functional food market. A wide range of customers (47%) opined that fortified foods and drinks satisfy their recommended nutritional requirements (Sloan, 2003). In this regard, fortified supplements in the form of blended drinks are a good option which has enhanced taste as well as improved nutritional value. Recently there has been an exceptional demand in the food industry for inexpensive proteins and bioactive peptides for human consumption. Several attempts have been made on utilization of protein hydrolysate in the formulation of various products but, still there is immense scope for its utilization in beverages especially in health-based energy drinks on account of its superior functionalities (Singh *et al.*, 2009). Additionally, alternative uses for co-products of the fish processing industry are highly sought-after as these co-products are excellent sources of nutrients like protein. The utilization of protein hydrolysate from cannery discards like tuna red meat for such health formulations is an ideal approach. The current study was conducted to formulate a health beverage incorporating protein hydrolysate from yellowfin tuna red meat.

Tuna protein hydrolysate (TPH) derived from yellowfin tuna red meat under optimum hydrolytic conditions was used. Based on RSM, 12 different ingredient combinations were prepared and subjected to sensory analysis to derive the best combination of health mix. The sensorily selected health mix was added with different levels of TPH *viz.*, 2.5, 5, 7.5 and 10% referred to as HM1, HM2, HM3 and HM4, respectively (Fig. 1). Health mix without addition of TPH was kept as control *viz.*, HM. Nutritional, functional, antioxidant, physical and sensory properties of the samples were assessed. Incorporation of protein hydrolysate in the HM improved the

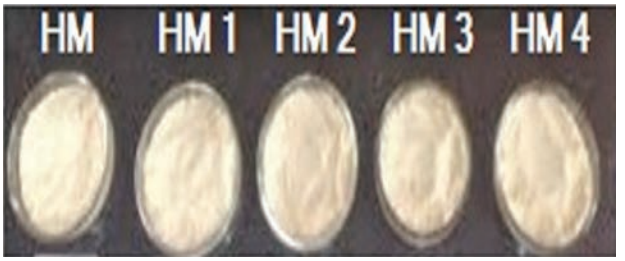

Fig.1. Health mix formulations with tuna protein hydrolysate

nutritional status *viz.*, protein, fat and ash contents of the health mix (Table 1). Moisture content varied from 5-5.6%. Addition of FPH improved the functional properties *viz.*, foaming capacity and emulsifying properties. Antioxidant property *viz.*, DPPH radical scavenging activity also revealed slight increase from 21.78% for HM to 26.64% for HM4. The colour parameters of the health mix indicated an increase in lightness and yellowness values, whereas a decrease in redness values were observed for samples containing higher levels of protein hydrolysate. Sensory studies indicated highest acceptability for control followed by health mix with lower level of protein hydrolysate. HM2 exhibited slight detection of fish flavor whereas it was prominent in HM3 and HM4 (Fig. 2). Studies revealed the potential of tuna protein hydrolysate for incorporation in health beverage formulation for its quality enhancement.

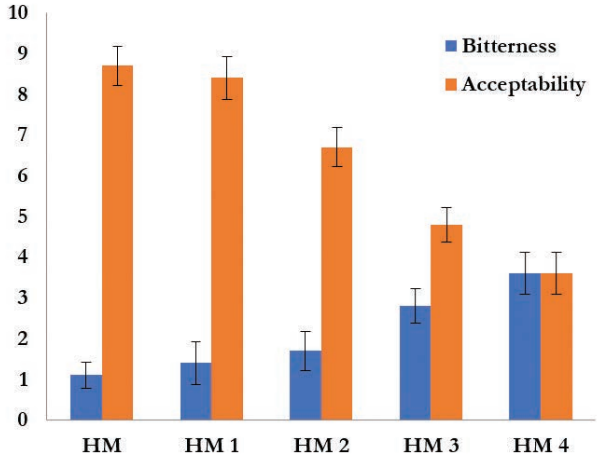

Fig.2. Variations in sensory properties of health mix samples

Table 1. Proximate composition of health mix samples

Sample	Moisture	Protein	Fat	Ash	Carbohydrate
HM	5.61±0.16	9.69±0.14	1.23±0.01	0.93±0.02	82.54±0.16
HM1	5.23±0.11	11.82±0.32	1.36±0.05	0.95±0.02	80.64±0.35
HM2	5.19±0.12	12.84±1.04	1.49±0.06	1.05±0.08	79.43±1.04
HM3	5.17±0.10	14.36±0.48	1.45±0.04	1.20±0.06	77.82±0.56
HM4	5.01±0.15	15.10±0.26	1.45±0.08	1.76±0.11	76.68±0.11

References

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Microwave vacuum drying: An innovative technology for rapid drying of fish

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Drying is an age old and least expensive method for preserving the quality of fish for a long period. Dried fish represent the cheapest source of concentrated protein to people in remote places, hilly areas and in locality where no water bodies are present. In India, 15-20% of the total fish catch is preserved by drying. Salt dried fish is highly demanded by the fish lovers because of its unique taste and has good demand in domestic and international markets. Traditionally, fish is dried openly under sun, which results in an unhygienic and low quality product. Over the years, several mechanical drying methods including hot air oven drying, combination of solar energy and mechanical drying, smoke drying, freeze drying, vacuum drying etc. have been developed. However, the fishermen still follow the unscientific practice of fish drying and the production of hygienic and export quality dried fish is being achieved only to a limited extend.

Microwave processing has been extensively used in the food industry for heating, cooking, pasteurization, sterilization and drying. Micro waves are electromagnetic waves of frequencies varying from 300 MHz to 300 GHz; smaller frequency waves having high penetration power. Microwave drying is a recent technique adopted for drying vegetables and fruits. In order to improve the drying rate and to enhance the quality of final products, other traditional methods are used in combination with microwave. Microwave vacuum drying has been used initially for drying vegetables like banana, carrot and potato slices. Through microwave vacuum drying, high product quality can be achieved by the low temperature and the rapid energy transfer of microwave heating. The combination of vacuum and microwave has the potential to reduce drying time, improve product quality and decrease energy consumption. However, the use of microwave-vacuum combination has not been much exploited for fishery products.

Visakhapatnam Research Centre of ICAR-CIFT has made an attempt to dry Indian mackerel using microwave vacuum drier. Indian mackerel was dressed in butterfly style and brined in common salt (4:1, fish to salt) overnight. Salted fishes were dried by microwave vacuum drying (MVD) and hot air drying (HAD). The moisture content of Indian mackerel was found to be reduced from an initial value of 76% to 32% by drying in a lab scale microwave vacuum dryer (600 W and 650 Hg mm) within 1.2 h whereas in hot air dryer (temp. 50-55 °C), fishes were dried for 12 h to reduce the moisture content to 32%. It is important to note that 10-fold reduction in drying time was achieved by employing microwave vacuum drying technique. Moreover, there was a marked difference between the colour and appearance of the fishes dried under both the methods where the MVD fishes scored higher than HAD fishes (Fig. 1). HAD fishes had salt particles condensed over the surface where as it was absent in MVD fishes. No significant difference was observed between the proximate composition of fishes dried by hot air and microwave vacuum drying. However, salt content in the muscle of MVD samples were higher than that of HAD samples.

Fig.1. MVD and HAD fishes

Rehydration ability and water absorption index of MVD samples were slightly higher to that of HAD samples. HAD sample had more salt-soluble and water soluble protein nitrogen fraction than MVD samples. Higher instrumental hardness values were observed for HAD samples. The study revealed that through microwave vacuum drying, drying time can be reduced to a greater extent compared to hot air drying. It was also realized that microwave vacuum drying can retain the original colour, improve the sensory attributes while maintaining the enhanced physico-chemical qualities. Microwave vacuum drying technique can be adopted by small scale and large scale entrepreneurs to market high quality dried fish products.

Molecular phylogenetic study of *femA* gene sequences of Methicillin Resistant *Staphylococcus aureus* in seafood

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Staphylococcus aureus is a common inhabitant of human skin surfaces and anterior nostrils of the healthy people and animals. But it's a well-known opportunistic pathogen that can cause a broad range of infections including mild skin infections, invasive diseases, and toxin-mediated diseases (Kroneberg *et al*., 2011). It is an ubiquitous gram-positive, catalase positive Cocci and facultative anaerobic bacteria and most frequently occurring food-borne pathogen world-wide by the presence of heat stable preformed Staphylococcal enterotoxins (FDA, 2012). Methicillin was introduced as a new β-lactam antibiotic in 1950 to overcome the penicillin resistant *S. aureus*. A decade later in 1961, Methicillin Resistant *Staphylococcus aureus* (MRSA) was reported in United Kingdom (Jevons, 1961) which is resistant to most recent β-lactam antibiotics (Katayama *et al*., 2000). In India, the significance of MRSA had been recognized relatively late and it emerged as a major health problem in the 1980s and 1990s (Mantri Rupali *et al*., 2014). MRSA is a major nosocomial pathogen causing significant morbidity and mortality (Sachdev *et al*., 2003).

Seafood contamination with antibiotic resistant MRSA could be a major threat to public health, as this resistance can be transferred to humans (Diana Gutiérrez *et al*., 2012). No strict guidelines are being practised in India regarding the use of antimicrobials in animal feeds as growth promoters that are used in human medicine (Patrick Butaye *et al*., 2003). The present study was undertaken to know the *femA* (factor essential for methicillin resistance) gene sequence differences in the identified MRSA isolates and its phylogeny in seafood (Sivaraman *et al*., 2017). The fish and fishery products (n= >400) were collected from the retail fish markets and fish processing establishments in Gujarat State, India and 19 number of isolates confirmed as *femA* positive.

A multiplex PCR was carried out with *mec*A gene (293 bp) and Staphylococcus genus specific (597 bp) for the confirmation of MRSA and found that 3.84% of the samples were positive for MRSA. These results made quite interesting to study the gene sequence of the factor essential for methicillin resistant gene A (*femA*) and was amplified by PCR (450 bp) as shown in Figure 1 and the DNA sequencing were out-sourced. 50 µl PCR reaction contains 200 µM dNTPs, 2.5 mM $MgCl_2$,

1X PCR buffer, 0.5U Tag DNA polymerase, 100 µg DNA/ µl with primer concentration of 0.6 pmol 16S rRNA, 0.8 pmol *femA*, 1.0 pmol mecA. The *femA* primer sequences of CGATCCATATTTACCATATCA and ATCACGCTCTTCGTTTAGTT.

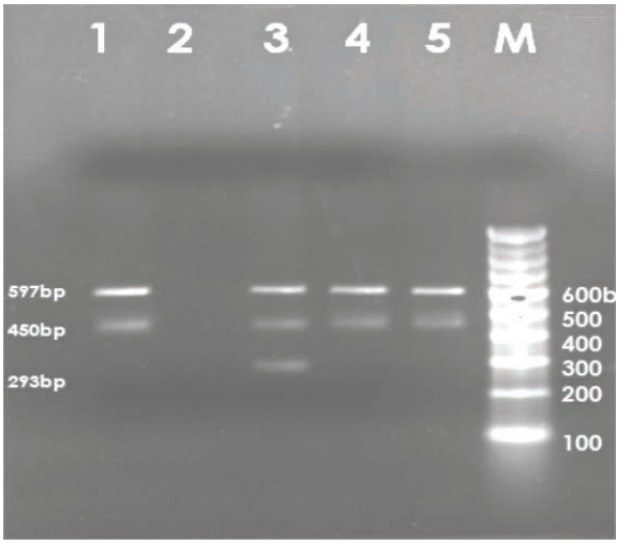

Fig. 1. Multiplex PCR amplification of MRSA isolates with genus specific primers (16S rRNA), femA and mecA gene (Lane 1: ATCC 25293 (MSSA) as reference strain; Lane 2: Negative control (without DNA); Lane 3: ATCC 43300 (MRSA) reference strain; Lane 4: Sample 1; Lane 5: Sample 2; Lane 6: 100 bp DNA ladder)

The phylogenetic tree was constructed based on the *femA* gene sequence differences from these 19 MRSA isolates by Clustal W (Weighted) method with the nucleotide substitutions of 1000 nucleotides and is shown in Figure 2. The DNA sequences were submitted to the ICAR DNA sequence submitting portal: http://webapp.cabgrid.res.in/dnadb/ (ID: 20150530051206, 20150530052426, 20150530052850, 20150530053351, 20150530053719, 20150530054137, 20150530054510, 20150530054804, 20150530055141,

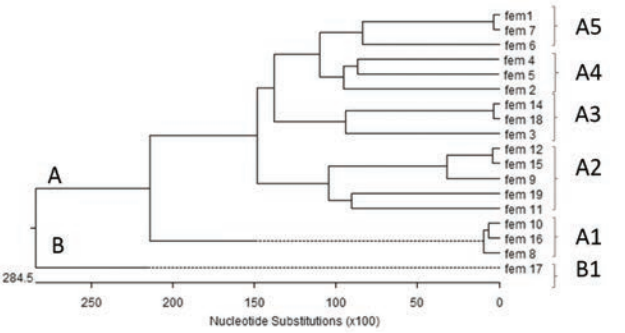

Fig. 2. Phylogenetic tree of the MRSA isolates based on femA gene sequence

20150530055802 and 20150530060133). Two distinct clade is formed based on the sequences and *fem 17* was alone in one clade (A) and the rest is in another clade (B). Whereas, in clade B consisting of two different subclade, the sequences of the isolate 8, 10 and 16 forming in one group (A1) and rest is on the another subclade (A2- A5). The formation of two distinct clades and also with significant differences among the A sub clade indicates the MRSA isolates may be from different source of contamination.

Hence the present study revealed that the presence of MRSA isolates in seafood is from the different sources i.e. possibly from infected fish handlers and processing and unhygienic environment of the fish source. Hence, all fish handlers should be made aware on the importance of personal hygiene and hygienic handling practices at all stages of processing, maintaining cold chain, adequate cleaning and disinfection of equipment and prevention of cross-contamination for ensuring the supply of safe seafood. This study highlights the need for continuous monitoring of antibiotic resistant pathogen MRSA in seafood with a view to prevent the source of contamination.

Marine origin *Bacillus* sp. : A potential collagenase source for fishery waste utilization

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Fisheries contributes immensely to global food and nutritional security. Substantial part of the fish produced were lost as fish waste discards and various other losses. Fish head, bones, gut, scales and skin are the major discard materials and anticipated to generate 32 million tonnes of waste from the processing operations (Kristinsson and Rasco, 2000). In this diction, utilization of fish waste is an important step. Among the discards collagen is a rich source found in skin, scales, and bones of over 7% of the total body weight of fish. Collagen is a major structural protein of animal origin which constitutes about 30% of total protein which is an insoluble fibrous protein. Among the enzymes highly marketed commercially, proteases takes the maximum share (Garcia-Carren˜o *et al.*, 1994; Arvanitoyannis and Kassaveti, 2008). Collagen hydrolysate prepared enzymatically using collagenase enzyme from microbial sources are comparatively better than thermo-chemically produced with strong alkali and high temperature (Rochima *et al.*, 2016).

In this context, a study was undertaken to screen seawater for isolating bacteria with collagenase activity/collagen degrading capability. Fifteen morphologically distinct bacteria were isolated from the seawater samples and identified both biochemically and molecularly using 16s rDNA sequencing analysis. All the 15 bacteria were characterized for the exo-enzyme activities such as lipase, protease, amylase and chitinase actives. Furthermore, the bacteria were tested for the collagenase activity with Azocoll Assay.

The study revealed that among the 15 bacterial strains isolated, 13 of them belonged to *Bacillus* sp. and two belonged to *Staphylococcus* sp. (Fig.1). Seven isolates were able to produce

Fig.1. Morphological characteristics of bacteria isolated from collagen medium

collagenase activity at 24 h of the growth and all the them belonged to *Bacillus* sp. (*Bacillus amyloliquefaciens, B. Velezensis* and *B. subtilis*) by Azocoll Assay. The sequencing study revealed the species *albeit*, there exist a variation in the biochemical characteristics which emphasize the potential novel species among the *Bacillus subtilis* group complex exempting *Bacillus velezensis*. The study also concludes the potential use of this enzymes in fish processing industry to convert waste to wealth.

Several attempts were made to isolate collagenase producing organism. However, all the attempts were confined to terrestrial ecosystem or food resulting in isolation of a few pathogenic organisms *viz.*, *Clostridium perfringens* and *C. histolytica, Bacillus subtilis* FS-2 (Nagano, 1999), *Bacillus subtilis* CN (Tran and Nagano, 2002), *Bacillus subtilis* AS1.398, (Rui *et al.*, 2009), *Bacillus pumilus* Co-J (Wu *et al.*, 2010), *Bacillus cereus* (Liu *et al.*, 2010) and *Streptomyces*

sp. Strain 3B (Petrova, 2006) and pathogenic *Vibrios*. The study reveals the potential use of this enzymes in the fish processing industry to convert waste to wealth and the possibility of new strain or species among the Subtilis group.

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Immobilization and sulphur oxidation capability of sulphur oxidizing bacteria

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Sulphur oxidizing bacteria (SOB) are one group of beneficial bacteria capabile of utilizing toxic form of sulphur produced by a group of bacteria called sulphur reducing bacteria (SRB) in natural environment during anoxic conditions (Friedrich *et al.*, 2001). The autotrophic SOB plays an important role in suphur cycle in maintaining the levels of hydrogen sulphide. The count of these bacteria in water and soil are usually very low and depend on the availability of the sulphur compounds in oxidized state. Natural polymers such as chitin, alginate, cellulose and chitosan are commonly used as carrier materials for immobilization of microorganisms where the bacterial cells are trapped. However, there is no such study on the immobilization of SOB. The present study is carried out to immobilize

the SOB using alginate and to characterize their oxidation potential.

Autrotrophic SOB was isolated from mangrove soils near Ernakulam, Kerala by employing Starkeys mineral salt medium (MSM) (Veerender *et al*., 2014). Out of 96 bacterial cultures, four isolates (SC-10, DN-5, SB-1 and SD-6) were selected for immobilization studies based on their pH reduction ability. Sodium alginate (100 ml) solution was prepared as per Smidsord and Skjak-Break (1990) with the addition of freshly grown SOB bacterial isolates (2 x 10^6 CFU ml^{-1}) into a strong cationic solution of 0.1 M strontium chloride solution (Himedia, Mumbai, India). Round beads formed in the solution are collected into a separate beaker with strontium chloride and left undisturbed for 1 h for complete hardening. The beads consist of the SOB trapped inside it. The beads were washed with sterile distilled water for several times and stored at 4 °C for further studies.

The sulphur oxidation potential of the beads was studied by inoculating the immobilized beads in various numbers (10, 50 and 100) in triplicates into 100 ml of MSM broth added with BPB to monitor the pH change. After inoculation the flasks were incubated at 30 ºC for a period of one week and monitored the pH change using a pH meter (Eutech Instruments, Mumbai, India). Sulphur oxidation capability of these bacterial isolates was measured in terms of sulphate ion (SO42-) produced in the medium by using spectrophotometer ($O.D_{450}$ nm).

When inoculated with 10, 50 and 100 number of immobilized beads, isolate SD6 showed maximum sulphate ion production (18.79, 19.04 and 21.95 mg ml^{-1}, respectively). The colour change is observed from blue to yellow during the growth of bacteria which is an indication of reduction in pH. Similarly, pH values are recorded in the range of 2.98 (Isolate SC-10; 100 beads) to 3.20 (Isolate SB-1; 10 beads). The size of beads ranged from 0.5 to 0.7 mm (Fig. 1). When the bacteria are immobilized by using suitable carrier material they have several advantages i.e. more tolerance to environmental conditions, optimized cell density, prolonged retention time and adequate protection from growth inhibitory substances such as antibiotics. Immobilized bacteria can also enhance the rate of sulphur oxidation which occurs slowly in normal conditions. Therefore, the use of immobilized SOB may be used as bio-inoculants in aquaculture farms to control the hydrogen sulphide toxicity thus providing a low cost management of eco-friendly technique.

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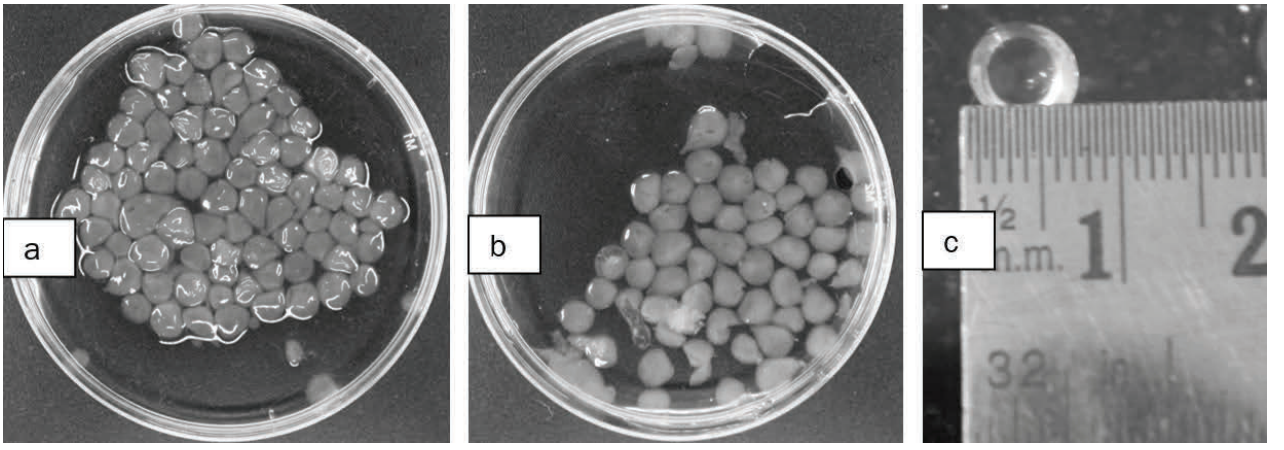

Fig. 1. Images of beads prepared with 3% sodium alginate. a) Control beads b) SOB immobilized beads c) showing size of bead

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"CIFTFISHPRO" - An information system on ICAR-CIFT value added fish products

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ICAR-CIFT, being a technology institute, has developed different types of value added fish products meant to enhance the livelihood of the society. These technologies are being transferred to the stakeholders through teaching and training. As a part of this, the knowhow about the technologies are printed in hardbound forms like leaflets, brochures, bulletins etc. As our country is progressing towards digital India, time has come to move from hardbound informations to digital information systems. This would allow the users to access the tangible information in their fingertips easily, irrespective of the location. By keeping this in view, ICAR-CIFT has developed "CIFTFISHPRO" - an information system on ICAR-CIFT value added fish products.

CIFTFISHPRO is an interactive information system with simple and easy navigation to access the contents. The system starts with an introduction about value added fish products, and further provides information on series of fish products. It gives an introduction about the product, information about ingredients required and step-wise method of preparation of the product. The series of fish products includes coated fish products like fish cutlets, fish fingers, fish burger, fish balls etc.; marinated products like fish and prawn pickles; extruded products like fish kure and noodles; wrapped fish products like fish momos, fish kebabs, fish samosa and fish rolls; cured products like dried fish and prawn and other products like fish sausage and prawn chutney powder. This also contains a contact form through which user can post any query by just mentioning name, e-mail id and content of the query; this would enable the experts from ICAR-CIFT to give reply to the query immediately.

Another interactive feature of CIFTFISHPRO is that the system will help the user to upscale the production of each product by automatically calculating the ingredients requirement for a given quantity of raw fish. The system also gives an option to the user to enter the input cost of the ingredients. This would give the user a broad idea about the total cost incurred while up-scaling the production of the product.

CIFTFISHPRO is available in the URL http://ciftfishpro.cift.res.in. This system has a home page, which contains product banners, an introduction, list of fish products, a contact form and contact us; the inner page contains list of fish products with a hyperlink to each product and up-scaling page for each product with a hyperlink. The home page of CIFTFISHPRO is developed in hypertext preprocessor (PHP) and hypertext markup language (HTML), the inner page is developed in HTML and up-scaling page is developed by using HTML and JAVA script. CIFTFISHPRO is a dynamic system as it can interact with the users and it can be updated at any time.

Recycle and reuse of abandoned fishing nets: Reports from Saurashtra, Gujarat

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Veraval Research Centre of ICAR-Central Institute of Fisheries Technology

Plastic is one of the wonderful materials which make our life better, easier and comfortable. This material significantly influences our life from dawn to dusk. But the problem created by the plastic to our environment is not much pleasant. As it is non-biodegradable, it sticks around for much longer than any other form of garbage. Estimates shows that approximately 80% of derbrises in the sea originate on land either swept in from the coastline or carried to rivers from the streets during heavy rain *via*. storm drains and sewer overflows. As per the estimate by Jambeck *et al*. *(*2015)., 275 million metric tons (MT) of plastic waste was generated in 192 coastal countries in 2010, with 4.8 to 12.7 million MT entering the ocean.

There are varities of articles which contribute to the plastic pollution in sea. Lost and discarded nets and lines from fishing vessels are one of the important contributors among them, especially in heavily fished areas (UNEP and NOAA). The abandoned and discarded fishing nets cause many problems throughout the world. These nets may be dumped on beaches, or sea shores which can create a navigational hazard for boats, or settle to the ocean floor to damage sensitive ecosystems. There are many reports showing that every year, tonnes of abandoned and lost fishing nets are dumped up on beaches. United Nations suggest that up to 10% of the trash that collects in our oceans is now comprised of this debris.

Proper disposal of the used net in fishing industry is necessary to reduce marine pollution and resultant ghost fishing. There are several initiative throughout the world to utilize such nets in several industries, including construction

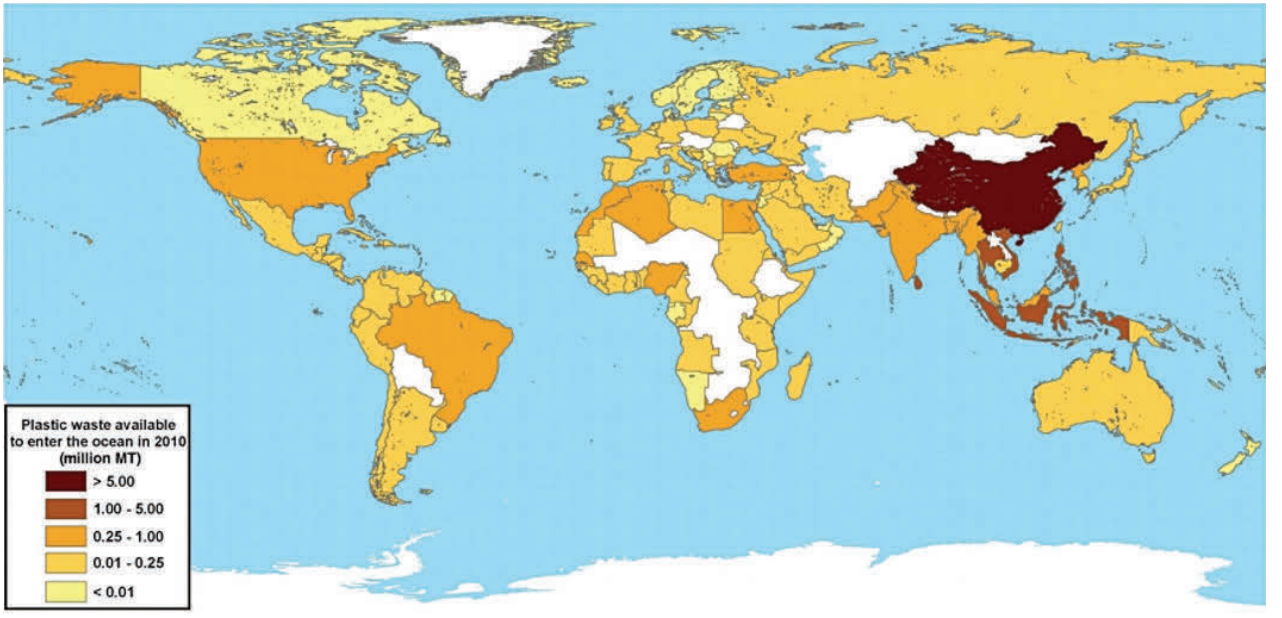

Fig 1. Global map with each country shaded according to the estimated mass of mis-managed plastic waste coast [millions of metric tons (MT)] generated in 2010 by populations living within 50 km of the (Source: Jambeck et al., 2015)

field (Ida Bertelsen, 2016). Nowadays, the idea to transform ocean waste into highly useable, recycled products is gaining attention around the world.

In the coastal region of Saurashtra, Gujarat there are several communities engaged in the collection and recycling of discarded fishing nets. The people engaged in this activity are travelling through the coastal districts mainly during off-fishing season. Recently, we have interviewed such a family from Porbandar, settled in Veraval, Gujarat. Presently four such families are there in the place where we conducted the survey. They put-up their tents and settle by the middle of May, and will stay up to the end of monsoon season. Fishermen carryout the maintenance and fabrication of fishing nets mainly during this season. The old nets are procured from the fishing villages and harbor by giving 300-500 INR per fishing net to the fishermen. The amount is fixed purely based on sensory evaluation, not based on weight or length of the net. Mono and multifilament nylon nets are mostly preferred for recycling, which are the materials used for the fabrication of gillnets. The big ropes used for tying the boats are also recycled. High Density Polyethylene (HDPE) is not much used for recycling. HDPE nets are normally used directly for fencing in agriculture fields and also used for other household purposes.

After procuring the net, they clean it properly to remove the foulers and borers. 7 am, and taken to an open space for drying. Later on unwinding of the yarn is done. After that, yarns will be passed through a machine and manual hand spinning will be done. This process is similar to the hand spinning of coir yarn. By this process, ropes with various diameters are prepared. In the case of monofilament nets, entire net is twisted suitably around a small rope (core) and made into long thick rope. In a season, more than 500 kg of fishing net is recycled by a family.

Once all the process completed, recycled ropes will be hanged near to their hut for sale. The

Data collection in progress

Collected net for recycling

Collected net

Rope making device

Unwinded yarns under processing

Rope made out of monofilament gillnets

Ropes ready for sale

rate of the processed rope will be in the range of ₹ 100-200/bundle. Price depends on quality and nature of material. A single unit/family earns ₹ 1000-2000 per month which provides subsistence to 10-20 nomadic families. Processed ropes are mainly used for agriculture purposes. Besides the recycling of fishing nets, they are also engaged in making of statues for festivals, knife sharpening, etc.

The recycling of fishing gear will help in reducing plastic pollution in the fishing sector and this will also reduce ghost fishing to some extent.

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Rapid detection kits (CIFtest Kits) to check adulteration of formaldehyde and ammonia in fresh fish

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Adulteration of fresh fish

Seafood is a promising food commodity with many beneficial health effects. Since fresh fish is highly perishable in nature, there is an emerging risk of economically motivated adulteration to enhance its shelf life which adversely affects consumer health. India's domestic fish market is reported to be selling formaldehyde adulterated fishes, especially in markets located far away from landing centres or production sites. According to Indian and International regulations, the fresh fish and shellfish should be preserved only by means of ice. Use of substance other than ice to extend the keeping quality is a fraudulent practice. Apart from direct application of adulterants, even adding ammonia-like substance during ice manufacture to slow down the melting of ice or to cut down the cost of ice, will also cause health problems to consumers. Even though, formaldehyde and ammonia are generated in very low levels in most living beings through normal metabolic activities, ingestion in large amounts through food can cause minor to serious health problems such as stomach pain, vomiting, coma, and even death. Formaldehyde is a potential cancer-inducing chemical which is a major health concern (IARC, 2004; Laly *et al*., 2018). Although ammonia is not so far reported to be carcinogenic, continuous ingestion of ammonia can lead to many health issues including injuries to mucous membrane of mouth, throat, esophagus and stomach. Ammonia readily dissolves in water and forms ammonium hydroxide, an alkaline solution. Ingestion of ammonium hydroxide can also result in corrosive damage to mouth, throat and stomach (Prajitna Sely, 2011). The use of prohibited substances in fresh fish can only be avoided by ensuring proper use of cold chain for fish and fish products during processing, storage, transportation and display for sale.

Rapid detection methods/ CIFTest

Continuous monitoring of these contaminants is time consuming and apart requires handling of huge quantity of toxic chemicals. The increasing consumer concern on food quality and safety issues has raised urgent demand for rapid, sensitive and portable screening methods. Taking these aspects into consideration, ICAR-CIFT has developed two rapid detection kits for checking adulteration of fresh fish with formaldehyde and ammonia. A special attention is given to develop a rapid detection technique as a non-destructive, simple and consumer-friendly method, to make its use easy for the domestic markets, and by general public. The kits consist of simple paper strips, reagent solution and standard chart for comparison of results.

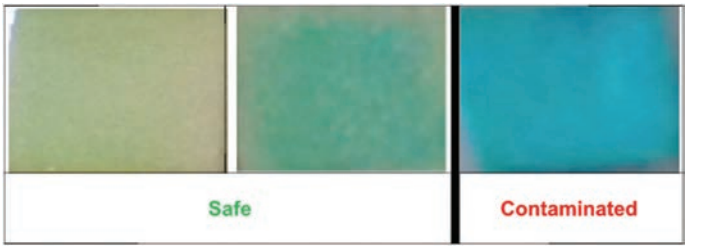
How to use the kits?

The paper strip is to be rubbed on the surface of the fish to be tested, and a drop of reagent solution is to be added to that. If the targeted contaminant is present in the fish, the paper strip gives a blue colour for ammonia and green to blue for formaldehyde within 1-2 minutes. The colour change can be compared with standard chart. The strips can also be used for meat (fillets and loins) of fish in the similar way to detect the presence of these contaminants.

Detection limits

The lower limit of detection of formaldehyde detection kit is 4 mg/Kg in case of fresh fish. Absence of formaldehyde adulteration in fish can result in slight yellowish colouration with a green

Formaldehyde detection kit for fresh fish



Ammonia detection kit for fresh fish

tinge. Fish adulterated with formaldehyde shows an immediate colour change within 30 seconds from a clear green colouration at lower level of adulteration (4 – 50 mg/Kg) to dark bluish colouration at higher level of adulteration (≥ 100 mg/Kg). The reagent solution provided along with the kit has to be used within 20 days of preparation. The paper strips has to be stored in safe container and should be kept away from moisture.

In the case of ammonia detection kit, the colour will be yellow at a level less than 100 mg/Kg to green at level ranging from 100-300 mg/Kg which comes under safe level of consumption. The development of blue colour in the strip indicates ammonia content of more than 300 mg/Kg and can be considered as adulterated.

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Infrared fish dryer

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Fish is considered as an important food stuff due to its high protein content and nutritional value. Fresh fish contains up to 80% of water. It is a highly perishable food material and has very short storage life. Drying is the most common preservation technique employed to increase the storage life of fish. It preserves fish by inactivating enzymes and removing the moisture necessary for bacterial and mould growth (Duan *et al.*, 2004). Traditionally fish drying is carried out by open sun drying method which is extremely weather-dependent and has problems such as contamination with dust, soil, sand particles and insect infestation. Therefore new drying techniques and dryers must be designed for hygienic drying of fish. Nowadays, various hot air dryers are being used for drying of food products. However, hot air drying has drawbacks of both long drying time requirement and poor quality (Chou and Chua, 2001).

In recent years, infrared drying has gained popularity as an alternative drying method for foods. Infrared is an electromagnetic wave which can penetrate into the interior of the food, where they are converted into thermal energy and providing a rapid heating mechanism. When infrared is used to heat or to dry foods, the radiation impinges on the exposed material surfaces and penetrates to create internal heating with molecular vibration of the material and the energy of radiation is converted into heat (Ginzburg, 1969). During drying with an infrared heat source, the energy in the form of electromagnetic waves is absorbed directly by the product without any loss to the environment leading to considerable energy savings. Infrared heating offers many advantages over conventional hot air drying. The use of infrared radiation technology for drying has several advantages such as decreased drying time, high energy efficiency, high quality finished products and uniform temperature in the product (Kocabiyik and Tezer, 2009).

ICAR-CIFT has designed and developed an infrared dryer for efficient drying of fish and fishery products. A 5 kg capacity prototype of the red dryer was designed using marine plywood, infrared lamps, heating element and stainless steel trays (Fig. 1). Energy requirement for drying was distributed by eight infrared lamps of 150 W each. Dryer was operated at no load and load conditions. Temperature attained in the dryer under no load condition was 60±0.5 °C within three hours of operation. Drying experiments conducted for fruits and vegetables also showed promising results.

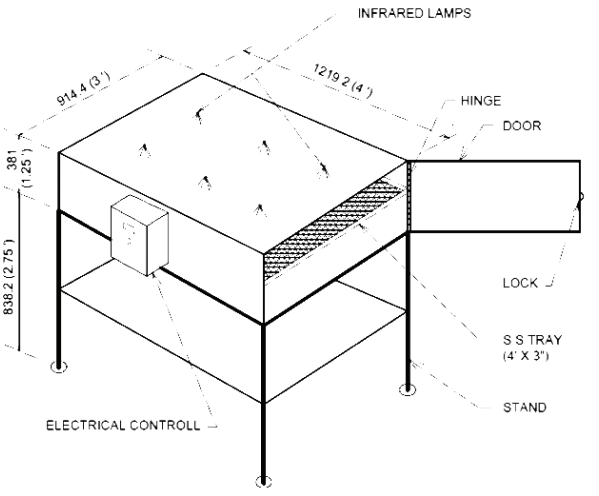

Fig. 1. Infrared dryer prototype

Drying experiments were conducted using different fish species and small shrimps. It was observed that the drying time requirement is less in infrared dryer than conventional electrical dryers. Under no load condition the drying temperature of 63 °C was attained in 0.5 h and the air velocity and relative humidity

Specifications of the infrared dryer prototype

Heat source	:	Electrical
Rating of infrared lamp	:	150 W
No. of lamps	:	6
Dimensions (l x b x h)	:	1.219 x 914 x 0.381m
Tray dimension (l x b)	:	0.9 x 0.60 m
Tray material	:	Stainless steel
Total drying time	:	4-5 h
Loading capacity	:	5 kg
Suitable items for drying	:	Fish and fish products, spices, vegetables, fruits and agro-products

was recorded as 2.5–2.8 m/s and 55–60%, respectively. Performance evaluation of the dryer was conducted using different fish species like, threadfin bream, sardine, shrimps etc. Moisture content of shrimp was reduced from 77 (% wb) to 10 – 12 (% wb) in 5 h of drying at drying temperature of 60 ^{o}C, air velocity of 1.5 ms^{-1} and relative humidity of 60%.

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Green synthesis of gold nanoparticles using different reducing agents of aquatic origin

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Nanotechnology exists among the most fastest growing branches in science and engineering because of the unique physical and chemical properties of nano materials, such as novel electronic, optical and magnetic properties, catalytic activity etc. These properties gave applications for nanoscale metals in biotechnology, material science and chemistry. Gold nanoparticles were already used for medical and staining purpose since 16th century Santhoshkumar *et al.,* 2017). The Foods and Drugs Association approved the nano therapeutic approach with nanoparticle-based anticancer drugs (Tiwari *et al*., 2011). Development of biosensors and biolabels using metal nanoparticles has recently gained extensive importance. Green synthesis of nanoparticles has several advantages over conventional methods involving chemical

agents associated with environmental toxicity. These include polysaccharide, mixed-valence polyoxo metallates, irradiation, Tollens (Ahmed *et al*., 2016) and biomolecules. Many of these molecules act as stabilizing agents, sometimes as both reducing agent and stabiliser. Several studies have been done on the development of metal nanoparticles using plant extracts (leaves and flowers) as they are rich in phyto-chemicals with aromatic hydroxyl, carboxylic and aldehyde functional groups. Chitosan, a biodegradable and bio-compatible molecule have been widely used as both protective and reducing agent. This study aims to develop gold nanoparticles using different sources or reducing agents.

The AuNPs were synthesised using 1Mm auric chloride at a temperature of 90 °C for 30 minutes. Chitosan, squid skin protein hydrolysate, carrageenan and shuck water from black clam were used for reducing gold chloride. The chitosan, squid skin protein hydrolysate and carrageenan were used at a concentration of 1% (w/v) and shuck water was used at 10% concentration (v/v). The collective oscillations of the electron cloud at the surface of AuNPs results in surface plasmon resonance (SPR) which is responsible for the colour exhibited by the solution (Regiel-Futyra *et al*., 2015). The surface plasmon bands (SPBs) for gold nanoparticle give characteristic UV absorption bands around the 500-550nm (Tiwari *et al*., 2011). The spectral bandwidth, peak position and intensity of the SPBs are attributed to

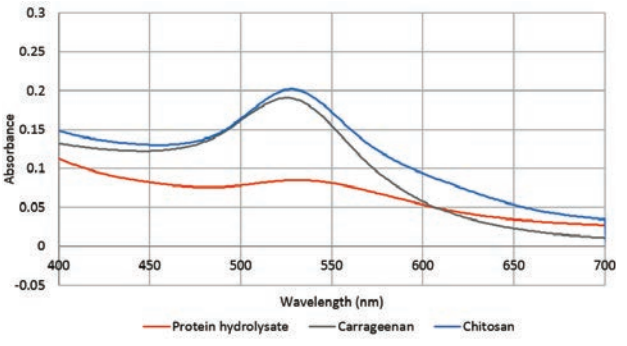

Fig. 1. UV visible absorbance spectra of AuNPs with squid skin protein hydrolysate, chitosan and carrageenan

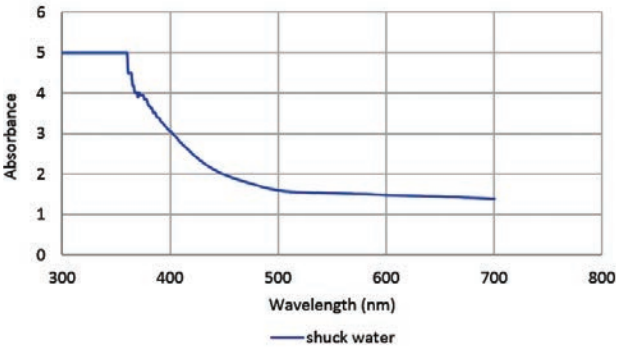

Fig. 2. UV visible absorbance spectra of gold chloride in shuckwater

AuNPs shape, size and productivity. The AuNPs synthesised with chitosan, squid skin protein hydrolysate and carrageenan gave typical band with peak at 520-550 nm (Fig. 1) and this proves the efficiency of these biomolecules to reduce gold ions to nanoparticles. The shuck water was not able to reduce the gold chloride hence there was no colour change of the solution and the characteristic band (Fig. 2) of AuNPs was absent.

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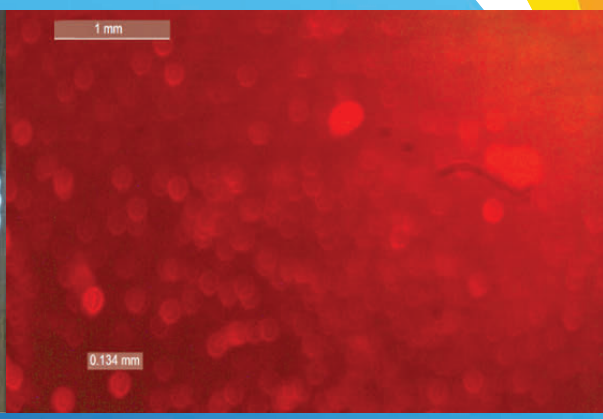

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