Research Highlights
2017-2018

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Dissemination of knowledge acquired through research is necessary for effective transfer of technology. ICAR-CIFT, being quite aware of this need, has therefore projected this publication highlighting the research activities undertaken during 2017-18 in a nutshell.

The year that went by, was a period of notable achievements for ICAR-CIFT which are summarized in this publication. Research activities undertaken in the fish harvest area include designing of deep sea fishing vessels under the Blue Revolution Scheme of Govt. of India; nano technological interventions in surface modification of boat building steel for corrosion resistance etc. Post harvest research activities carried out during the previous year mainly focused on the development of novel processing technologies and value addition from emerging farmed fish species, unconventional fishery resources and efficient utilization of fishery waste such as freeze chill technology for pufferfish, bio-silaging of fish waste using sugarcane etc. Seafood value addition viz., smoke flavoured chips and fingers from Nile tilapia, instant battered and breaded product mix, ethnic RTE fish products etc. were developed. With special emphasis to seafood quality aspects, research towards the development of oxygen scavengers, freshness indicators etc., were done. Development and validation of paper strip-based rapid detection kits for checking ammonia and formaldehyde adulteration in fish was one of the major research achievement during the year. Validation of cold fill and hold process in acidified fish and prawn pickle; collection of geo-referenced data on Cephalopods to identify hot spots of cadmium in coastal fishing grounds; monitoring of foodborne pathogens in fish and shellfish species from markets and harbours; analysis of samples procured from Kerala, Karnataka and Gujarat region for the presence of ciguatoxin; isolation of \textit{Thiobacillus aquaesulis}, a potential sulphur oxidizing bacteria; metagenomic analysis of aquaculture farm samples for ammonia oxidizing bacterial diversity, identification of three potential qorum sensing \textit{Bacillus} spp. as an alternative to antibiotics to protect \textit{Peneaus monodon} post-larvae from \textit{Vibrio harveyi} infection were some of the research activities undertaken for monitoring the safety aspects of fishery sector. Research highlights in the biochemical and nutritional aspects of seafood viz., development of \textit{thiamine} and \textit{pyridoxine} loaded vannilic acid-grafted chitosan and pectin-based multiple emulsion as a potential delivery system for squalene; extraction of fucoxanthin and lipid from brown seaweed by super critical fluid extraction method; extraction of a marine biopolymer, carrageenan from red seaweed; development of a pain relieving balm with squalene as an active component and a skin moisturizing lotion with collagen, chitosan and alginate as main ingredients were done. Remarkable achievements were made in fishery engineering area by development and commercialization of various machineries such as modern, hygienic and refrigeration-enabled mobile fish vending kiosk; development of energy and cost-efficient infrared dryer for fish; development of a Peltier-based 12 V battery-operated specimen collection and transport cooling device of 5 L capacity etc. Studies leading to the identification that the perceived strength and weakness of the fish entrepreneurship development are family support and marketing of traditional fish products and administrative and legal hurdles as the major threats; identification of nodes and actors prevailing in the value chain of reservoir system; documentation of ITKs relevant to the fisheries sector, specifically with reference to fishing and oceanographic parameters, fish shoal identification, fishing craft and gear and fish processing etc. were also carried out during the period.

I hope that this publication will be of interest to all those who are directly or indirectly related to fish and fisheries, academicians, scientists, research scholars, fishermen and entrepreneurs.

Kochi
6th September, 2018

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Director
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- Surface modification of boat building steel was done by using nano titanium oxide-cerium oxide-manganese dioxide mixtures in different combinations. The results showed 0.01:0.005:0.005 MnO:CeO:TiO combination with good corrosion resistance.

- Multi-locational field evaluation of nano copper oxide coated PE-PANI aquaculture nettings showed excellent fouling resistance and the technology is ready for commercialization.

- Deep sea gillnet trials revealed maximum catch in nets rigged at 0.5 (53.16%) followed by 0.4 (23.56%) and 0.6 (23.26%) hanging ratio. Among different modes of capture, wedging and entangling (37.26%) and wedging alone (29.61%) contributed the most to the catches.

- Associated with M/s Cochin Shipyard Ltd. for design development of deep sea fishing vessels under Blue Revolution Scheme of Sagarmala.

- Signed MoA with Department of Fisheries, Andaman and Nicobar Administration: (1) to design up to 19 m FRP fishing craft for fishermen in the Andaman and Nicobar islands under the centrally sponsored scheme, and (2) to design 7.6 m to 14 m fishing crafts in various sizes under the Blue Revolution Scheme.

Exposed untreated and treated netting

Percentage of fish caught by different mode in deep sea
Comparison between ‘J’ and ‘Circle’ hooks has shown that in both types of hooks, the pattern of hooking was almost same and in the case of ‘Circle’ hook, lower lip hooking was 12.5% while in ‘J’ hook it was 6.25%. In ‘J’ hook, 10.7% fishes caught had severe bleeding while no fishes caught by the ‘Circle’ hook recorded severe bleeding. Post-release survival was 100% in fishes caught by both type of hooks.

Comparitive studies and catch characteristics of the low drag and HDPE fish trawls were carried out. The average CPUE of the low drag and HDPE fish trawls were 17.34 kg.h⁻¹ and 10.58 kg.h⁻¹, respectively. Though the CPUE was higher in the low drag trawl, the species composition of the catches were not significantly different as manifested by multivariate tests like Multi-Dimensional Scaling (MDS) and ANOSIM.

Retention and exclusion characteristics of fish species at trawls attached with 40 mm square mesh codend at Bay of Bengal was assessed. Of the total fish catch 81.4% was retained in the trawl codend and 18.2% was excluded. Overall catch excluded with 40 mm square mesh codend, during the period of observations was about 17.8% of the total catch.

The selection properties of *Johnius carutta* in 40 mm square mesh codend was worked out using covered codend method. The L50 value was worked out as 13.8 cm and the optimum mesh size for use in the codend was estimated as 50.6 mm.

Retention and exclusion characteristics of fish species at trawls

The effects of freeze-chill technology and noni (*Morinda citrifolia*) fruit extract on the shelf life of pufferfish indicated two days of shelf life extension in treated samples compared to control samples under chilled condition.

Incorporation of dietary fibre (wheat fibre, oats fibre or psyllium fibre) in threadfin bream (*Nemipterus japonicus*) sausage indicated positive results for oat fibres.

The benefit-risk ratio of consuming
Ready To Eat (RTE) tuna products indicated low risk of consuming yellowfin tuna in TFS cans and retortable pouches.

- The cage-reared Nile tilapia showed higher overall acceptability compared to pond-eared counterpart, in terms of colour, flavor, and appearance. Bleeding of Nile tilapia prior to storage improved colour and functional properties of proteins.
- The use of magnesium chloride along with sodium chloride (0.45%) in the range of 0.1-0.2% in wash water yielded surimi with better textural properties.
- An instant mix for battered and breaded fishery products using fish flour was developed with satisfactory rehydration capacity and good textural and sensory acceptability.
- Bioplastic films from PLA manufactured by incorporating different clays like Montmorillonite, Halloysite and Bentonite at different levels were found suitable for packaging of tilapia.
- Polylactic acid films incorporating cellulose nanofibers at different levels (1-5%) were found to be suitable for chilled storage of flat head mullet (Liza parsia) fish.
- Chill storage studies of milkfish (Chanos chanos) in palm sheath trays with shrink and stretch overwrap demonstrated palm sheath trays as an ideal biodegradable material for retail packaging of fish in low temperature storage conditions.
- Use of iron powder with 200 mesh size enhanced the efficiency of oxygen scavenger compared to 120 mesh iron powder.
- Gold nano particles synthesized using chitosan can be used to distinguish freshly packed products from frozen stored products.
- Chill storage of cuttlefish (Sepia pharaonis) skin enhanced the susceptibility towards proteolysis with increase in storage period.
- A modified method of bio-silaging of fish waste using sugarcane waste as the source of sugar was developed.
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- Developed and characterized seaweed extract-based bio-plastic.
- Developed and characterized seaweed extract-based biodegradable suture.
- Developed a technology for extracting seafood flavour peptides from shrimp.
- Prepared gelatin hydrolysates having promising antioxidant and functional properties from the skin and scale of solefish.
- Developed a technology for making dried fish fingers from tilapia mince.
- Standardized microwave vacuum drying technology for drying fish, shrimp and squid shreds, with superior quality products.
- Developed technology for making gravads and smoke flavoured steaks from Nile tilapia.
- Studies on the effect of 5-MeV electron beam (0, 2.5, 5.0 and 7.5 kGy) irradiation and vacuum packaging on the shelf life of headless vannamei (Litopenaeus vannamei) stored at 2 °C indicated that control and 2.5 kGy treated vannamei had a shelf life of up to 12 days and 14 days, respectively while 5.0 kGy and 7.5 kGy treated samples were rejected on 28th day.
- Studies on the effect of vacuum packaging and E-beam irradiation on the quality and shelf life of peeled vannamei during chilled storage (2 °C) indicated that control, 2.5 kGy and 5.0 kGy treated peeled vannamei had a shelf life of 10 days, 13 days, 18 days, respectively while 7.5 kGy treated sample was rejected on 23rd day.
- Shelf life study of Modified Atmospheric Pressure (MAP) packed chill stored headless vannamei shrimp and seerfish steaks indicated a shelf life of 10 days.
days for vannamei and 14 days for seerfish.

- Evaluation of the efficacy of bulk zinc oxide in combination with chitosan against seafood pathogenic and spoilage bacteria indicated it to be a suitable alternative to be added to seafood products to control pathogenic and spoilage bacteria.

### Biochemistry and Nutrition

- Squalene powder developed had oxidative stability of four months.
- Pectin-based multiple emulsion was developed as a potential delivery system for squalene and anthocyanin.
- Supplementation of fish oil rich in squalene modulates the mRNA and protein expression of enzymes of lipid metabolism.
- Chitosan nano particles were prepared by ionic gelation method for the entrapment of anthocyanin as an effective strategy to enhance their in vivo bio-availability and in vitro stability. Oral supplementation of anthocyanin-loaded nano particles were found to have hypolipidemic effect in high fat-alcohol fed experimental rats and also protected gastric mucosa against HCl-ethanol induced damage.
- Thaimine and pyridoxine-loaded vannilic acid-grafted chitosan was developed and its anti-inflammatory effect in male Wistar strain albino rats was evaluated.
- Bioactive collagen peptides were prepared through enzymatic digestion of acid soluble fraction of skin collagen from hammerhead shark (*Sphyrnae mokkaran*) followed by subsequent column chromatographic fractionation with 94% radical scavenging activity.
- Omega-3 and Omega-6 fatty acids extracted using supercritical fluid extraction from sardine gave better yield and quality of fatty acids when compared to conventional extraction methods.
- A pain relieving balm with squalene as an active component was developed. *Cissus quadrangularis* extract was added as an additional component which has healing property in bone ailments.
- A skin moisturizing lotion was developed with collagen, chitosan
and alginate as main ingredients. extracted by supercritical fluid extraction method.

- A marine biopolymer, carrageenan was extracted from the red seaweed *Kappaphycus alvarezii*, by hot alkaline water treatment and characterized by FTIR Spectroscopy.

- Characterized and evaluated seaweed waste as a suitable feed ingredient in fish feed industry.

- Fucoxanthin and lipid from brown seaweed (*Sargassum* sp.) was

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**Quality Assurance and Management**

- Validated the cold fill and hold process in acidified fish and prawn pickle for 5 log reduction of vegetative bacterial pathogens such as *Salmonella enterica*, *Listeria monocytogenes* and *Escherichia coli* O157:H7 as required in Code of Federal Regulations (21 CFR 114).

- PCR technique was found to be better option for the recovery of food-borne pathogens such as *L. monocytogenes* and *S. enteritidis* using 3-tube method in fish matrix, when compared to conventional plating method.

- Geo-referenced data on Cephalopods (n=36) were collected
Studies on bio-accumulation of cadmium in various tissues of squid (muscle, digestive glands and tentacles) showed highest concentration of 8.79±0.1 ppm in the muscle tissue of *Uroteuthis singhalensis*.

- Food-borne pathogens recoveries from different food matrix indicated 65%, 54%, and 60% recovery from raw fish, shrimp and squid samples, respectively.
- Monitoring study of food-borne pathogens in 30 species of fish and shellfish procured from Thevara fish market and Cochin Fisheries Harbor showed that mesophilic bacterial load was 5.38-8.11 log CFU/g.
- Paper strip-based rapid detection kits for checking ammonia and formaldehyde adulteration in fish was developed with LoD of 4 ppm and 350 mg/Kg.

**Rapid detection kit for formaldehyde adulteration**

- Raw, salted and dried Leather Jacket (*Scomberoides* spp.) from Navabandar Region of Gujarat were free from pathogenic bacteria though higher yeast and mold count (1.2x10³ CFU/g) in salted and dried samples were observed.
- CO₂ and N₂ ratio of 60:40 in MAP were found to extend the shelf life of *Scomberomorus commerson* (Seerfish) steaks by 8-9, 5-6 and 2-3 days, compared to air packed LDPE, laminated pouches.
- Water activity of 6.81% of salted and dried fishery products (n=40) was found to be above the FSSAI prescribed limit of 6.81% and salt content was less than 12% in 26.67% of samples.
- Fermented fishery products sourced from Tripura were free from all major food-borne pathogens, but arsenic was detected at trace levels (1.0 -1.9 ppm) in ‘Puthi shidal’ and 0.8 - 1.2 ppm in ‘Phasa shidal’.
- Hygiene status of three fishing harbours and one aquaculture farm revealed the presence of hygiene indicator bacteria to a varying degree and water available for use as major source of contamination.
- Ciguatoxin was not detected in fish samples (n=26) procured from Kerala, Karnataka and Gujarat region.
- Analysis of fish pickles from various origins revealed that the pH were in the range of 4.00-4.30, aₜ was
Salmonella was present in 3.3% of dry fish in Kochi region. *Salmonella* Typhimurium, *Salmonella* Urbana, *Salmonella* Paratyphi B and *Salmonella* Salamae were the serotypes of *Salmonella* found in seafood samples of Kochi.

1/2a, 3a, 1/2b, 3b and 7 are the serogroups of *Listeria monocytogenes* found in seafood samples of Kochi.

*Vibrio harveyi* was found in 7.8% of hatchery and farm samples.

t334, t311, t304, t3481 and t127 spa types of MRSA were found in landing centers and retail markets of Kottayam district, Kerala.

*Thiobacillus aquaesulis*, a potential sulfur oxidizing bacteria isolated from the aquaculture farm was optimized for immobilization studies.

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<td>A total of 132 samples screened for heavy metals showed the presence of arsenic (BDL-16.2 ppm), cadmium (BDL-1.41 ppm), and lead (BDL-0.209 ppm), respectively.</td>
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<td>Pictorial guidelines were prepared for 16 species of fish which included farmed freshwater fish, farmed brackishwater shrimp, wild marine shrimp, cuttlefish and marine fish for on-site examinations for spoilage by the customers.</td>
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**Microbiology, Fermentation and Biotechnology**

Metagenomic analysis of samples for ammonia oxidizing bacterial diversity revealed that Phylum Chloroflexi was a dominant Phylum followed by Proteobacteria in aquaculture farms.

Nine tyrosinase producing Actinomycetes were isolated from aquatic environment.

Three potential quorum quenching *Bacillus* were identified to protect

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Penaeus monodon post-larvae from Vibrio harveyi infection.

- Coagulase negative Staphylococci viz. Staphylococcus warneri, S. haemolyticus, S. xylosus, S. simulans, S. auricularis and S. epidermidis were identified from seafood of Veraval, Gujarat.

- Cronobacter sakazakii was isolated from 10 seafood and environment samples screened.

- Draft genome sequence of two methicillin resistant Staphylococci (MRSA) isolate (ST 1 and ST 39) from the salted dry fish from Gujarat was completed.

- Three samples were found to be positive for WSSV out of 48 shrimp samples.

Engineering

- Modern, hygienic and refrigeration-enabled mobile fish vending kiosk was designed, developed and commercialized.

- Redesigned the existing ICAR-CIFT solar-LPG hybrid dryer with notable innovations.
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- Performance evaluation of the newly designed multi-purpose (Fish drying, water heating and electricity) solar thermal conversion system with biomass heater backup was carried out.
- Energy and cost efficient infrared dryer for fish was designed and developed.
- Performance evaluation of ICAR-CIFT hand operated fish descaling machine was conducted to optimize operating conditions for various fishes.
- Developed Peltier-based 12 V battery-operated specimen collection and transport cooling device of 5 L capacity.

Infrared drying of shrimp

Descaling efficiency under various loading conditions after nine minutes of descaling

Extension, Information and Statistics

- The perceived strength and weakness of the fish entrepreneurship development are family support and marketing of traditional fish products, respectively in the selected study area. Administrative and legal hurdles are the major threat. The opportunity is the Government funded livelihood schemes for the fisherfolk on entrepreneurship development.
- The Entrepreneurial Intention (EI) for the fisheries enterprise was influenced by personal attraction, perceived behavioral control and professional option.
- Seven nodes and actors prevailing in the value chain of reservoir system were identified. And for the economic valuation, both the use and non-use values of ecosystem were also identified. PESTLE analysis was carried out towards identifying the concerns and opportunities in the reservoir fisheries.
- The K-co-efficient and VPA