Marine Fisheries Information Service Technical & Extension Series

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### Marine Fisheries Information Service Technical & Extension Series

#### **Marine Fisheries Information Service**

Technical & Extension Series

Mar. Fish. Infor. Serv., T & E Ser., No. 246, 2020

#### Published by

Dr. A. Gopalakrishnan Director ICAR - Central Marine Fisheries Research Institute, Kochi

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Sea cage farming for cobia in Gulf of Mannar (Photo credit: Mandapam Regional Centre)

Marine Fisheries Information Service Technical and Extension Series envisages dissemination of information on marine fishery resources based on research results to the planners, industry and fish farmers and transfer of technology from laboratory to the field.

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#### From the Editorial Board

Warm greeting to all our esteemed readers

As apex predators in the oceans, marine mammals play a key role in the marine food web dynamics and ecosystem balance. As they are affected by fishery (by-catch in fishing gears, collision with fishing vessels) and non-fishery (climate change, hunting, habitat degradation) factors, their conservation and management becomes complex. Globally, species specific database on marine mammals is scanty and the lead article in this issue of MFIS, takes a look at the taxonomic issues of marine mammals in the Indian EEZ which is vital for formulating their stock management strategies. Seaweeds are highly valued marine resources traditionally used as food in several Asian countries. They are also used as raw materal for the extraction of phycocolloids, critical inputs for several industries including pharmaceutical, cosmetic and food processing. Seaweed farming is to be faclitated by mapping of the potential sites which is highlighted in a recent study conducted in all maritime states of India . Open sea cage farming of cobia has become very popular among fish farmers, especially on the south east coast of India. Hence the focus shifts to healthy animal husbandry practices and disease prevention in aquaculture systems which is detailed in an article on fish vaccines developed and tested in cobia cage farms in Gulf of Mannar. In the face of the COVID 19 pandemic that played out in the year 2020, marine fisheries sector faced numerous challenges and it is reassuring that the stakeholder community successfully addressed most of these concerns. Moving forward, we extend best wishes to all our esteemed readers for a Happy New Year 2021.



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# Taxonomic identification of marine mammals – current research and approaches

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#### Introduction

The term 'marine mammal' includes members of 5 different mammalian groups: cetaceans (whales, dolphins, and porpoises), sirenians (manatees and the dugong), pinnipeds (sea lions, the walrus, and seals), marine and sea otters, and the polar bear. They are warm-blooded animals which have undergone major adaptations that permit them to live in water. These involve the loss of hind limbs (cetaceans and sirenians). the adaptation of limbs for propulsion through water (pinnipeds), and the general streamlining of the body for hydrodynamic efficiency (all 3 groups). Structural modifications to the sea otters and the polar bear are less apparent in body form and they continue to closely resemble their terrestrial counterparts. While cetaceans and sirenians spend their entire lives in the water, other marine mammals come ashore for various reasons, at particular times in their life cycle. In recent years, there has been a marked rise in the number of wildlife enthusiasts taking to educational and adventure expeditions to see marine mammals up close in their natural habitats. There is also increasing awareness of the integral importance of marine mammals to healthy aquatic ecosystems, and of the growing threats that a variety of anthropogenic activities, such as destruction of habitats, fishery interactions (e.g. gill net fishery), illegal fishing methods and pollution which challenge these animals and their environments. Research and education programmes should understand and clearly communicate these threats and recommend appropriate actions needed to reduce or eliminate their impacts.

## Current status of identification of marine mammal species

Accurate taxonomy is fundamental to the conservation efforts of living resources; the units on which conservation is based are determined partly by population structure and ultimately by species designation. Imperfect taxonomy may result, at least as much as a lack of understanding of the population structure, in the loss of genetic variability, e.g. unwitting extinction of a species. In cetaceans, morphological features are often subtle and difficult to compare because of the rarity of specimens or widespread distributions. A series of adult animals are required for the documentation of geographic morphological variation and such series may take decades to accumulate in museums and research institutions, unless large-scale fishery mortality accelerates the process. Thus identification of the geographical variants of recognized species of delphinids and phocoenids is difficult using the conventional approaches. Gaps in our present understanding of species status and geographic variation of cetaceans means that the list of currently recognized species of cetaceans will probably undergo serious revisions.

The order Cetacea comprises two extant sub-orders and one extinct sub-order. The extant sub-orders are Mysticeti- filter feeding (baleen whales) and Odontoceti (toothed whales) with at least 70 species, 40 genera, and 10 families. Both Mysticetes and odontocetes are thought to be descendants of Archaeocetes (Archaeoceti, ancient whales, known only from fossil records), an extinct sub-order. The number of extant species of cetaceans remains debated, ranging from 78 to 85. With the recent consensus that recognizes three rather than one species of Right whale (*Eubalaena* sp), the total number of species comes to 85, and the number of subspecies is reduced to 41. Recently one special issue of a dedicated journal for marine mammal research (*Marine Mammal Science*, 2017, 33) focussed only on species, subspecies and populations of cetaceans. Marine mammals are identified through morphologybased, photo identification and molecular taxonomy approaches by researchers.

Morphology based approach: Cetacean specimens "in hand" can be identified by using the dichotomous keys to external features. Conventionally, characters, such as ratio of the outer margin of the flipper to the total body length, colouration pattern, teeth count, comparative osteology, etc. are used to identify the cetaceans. Skulls of many species are sufficiently similar that only examination of a full series of each can define reliable diagnostic features. Great variability in morphological characters of cetaceans is not uncommon. Sometimes it may only be possible to label an animals or group as "unidentified long-snouted dolphin" or "unidentified beaked whale", etc.

A study of the available material in various museums and private collections before expanding the already reported number of species to a final inventory can eliminate the possible repetitions and bring out unknown details of a species. The need for more of the world's cetacean collections in museums and other institutions to be catalogued and accessible in digital mode is highlighted. This effort is already underway in many major museums, but the smaller collections remain relatively unknown. To facilitate access and comparisons, catalogues should ultimately be linked, managed and the information standardized through a single centralized location with the following data: collection locality and date, age/sex class, material collected (including soft tissue samples), total length and photographs of external appearance and skull morphology.

*Photo identification:* Photographs of dorsal fins and flukes help in identification of individual cetaceans and this technique, known as photo-identification, is useful for studying the school structure and species composition. A repeated photo-session from the same geographical location for a protracted period of time will help in monitoring resident and migrant populations as well as the reproductive success. Identification of the species at sea is quite different from that of a dead animal on land. Even under ideal conditions, an observer often gets little more than a brief view of a splash, blow, dorsal fin, head, flipper, or back, often from a great distance. Rough weather, glare, fog, or other bad sighting conditions compound the problem. Many species appear similar to another, especially in the brief glimpses typical at sea and a fair amount of experience and expertise to master the technique of identifying free ranging marine mammals at sea is necessary.

Generally, sightings from the survey boats are initially identified as "possible" or "confirmed" or as "unidentified", usually for the animals far away. Photo and video documentation of these sightings help to confirm the identification with the assistance of experts later. Sixty eight per cent of individual cetaceans sighted during one souther ocean cruise could be identified to the species level (Jayasankar *et al.*, 2007). Reports on vessel-based surveys to identify cetaceans based on their sightings are available from Maldives, Kerguelen islands, Mauritius, South China Sea, Mauritius to the Philippines, Indian Ocean, Seychelles, Caribbean Sea, Gulf of Mexico and Eastern Antarctica.

Molecular taxonomy: This approach must be firmly anchored within the knowledge, concepts, techniques and infrastructure of traditional taxonomy and is especially relevant for cetaceans, because (i) they are very mobile and inaccessible organisms for which morphological, physiological and behavioural characters can be exceedingly difficult to score for population studies and (ii) their highly derived and specialized morphology reduces the utility of phenotypic data for assessing their phylogenetic position within mammals. In general, molecular taxonomy outscores morphological taxonomy in the identification of groups showing little evolutionary differentiation, cryptic members of species complexes, members of closely related species that can only be identified at a particular life stage, inter-species hybrids, as well as in issues involving illegal fishing and marketing of endangered species. Illegal trade in animal/plant products is commonly practiced in some of the Asian countries, where they market some of the endangered species in the guise of ones approved by authorized bodies such as, the International Whaling Commission (IWC). Through a series of reports, IWC has published techniques and incidences of identification of market samples of cetaceans using DNA sequence analysis which has thus become a powerful tool for conservation by identifying the source of samples thought to be derived from threatened or endangered species. Only minute amounts of DNA are required, allowing for remote sampling. It is possible to use hair, blood, feces, skin biopsies and sloughed skin as a DNA source and the PCR-based techniques are simple and rapid, making them practical for conservation and population studies. In cetaceans and dugongs, the technique can be effectively used in the forensic identification of commercial products, verification of trade records and also for identifying ambiguous beach-cast specimens.

The rapid advances in molecular techniques of the past few decades have led to significant contributions towards improving cetacean taxonomy. At higher taxonomic levels, the increasing case of generating useful molecular genetic data, notably DNA sequences, paralleled by theoretical advances and the development of computer programs, has stimulated reinvestigation of phylogenetic issues involving cetaceans. In some cases, these investigations have led to revisions of taxonomic relationships. Molecular genetics can also aid taxonomic understanding of inter and intra-specific variations for conservation and management purposes. Mitochondrial DNA (mtDNA) is well established and widely used tool for species identification and to a lesser extent, population identification. Mitochondria are structures within cells that convert the energy from food into a form which cells can use. Although most DNA is packaged in chromosomes within nucleus, mitochondria also have small amount of their own DNA. This genetic material known as mtDNA spans about 16,500 DNA building blocks (base pairs) representing a fraction of the total DNA in cells. MtDNA is often used in studies of marine mammals for a number of reasons including its high rate of evolution, maternal inheritance, low effective population size and lack of recombination. It has helped to define management units for the effective management of the exploitation of any species.

Of the total of about 37 genes and non-coding regions in the mtDNA, one gene (cytochrome *b*) and a noncoding segment (control region) are most commonly used for studies on marine mammals. This is based on the advantages of their rapid evolution rate and variability which would facilitate accurate delineation of species and detection of population differentiation. DNA sequences from the control region and cytochrome b are reconstructed to develop a "tree" which would give clue to the exact or possible identity of the species. Molecular identification of marine mammals can be done in two steps: (1) sequence similarity search under BLAST (Basic Local Alignment Search Tool) as implemented in GenBank (www.ncbi.nlm.nih.gov) (2) once it is confirmed that the tissue sample originates from a cetacean, the species identity is searched within DNA Surveillance (www.cebl.auckland.ac.nz:9000/). All sequences in DNA Surveillance are included only if the specimen had been expertly identified and diagnostic skeletal material or photographic records were collected. The purpose of checking the higher taxa of the unknown sample with BLAST search is important because if it does not belong to the order Cetacea, results of the phylogenetic identification could be misleading.

# Studies on marine mammals in India

The Ministry of Earth Sciences (MoES) had funded ICAR-CMFRI to study biology, trophodynamics, fisheries interaction, contaminant accumulation, molecular taxonomy and PCR-based sex identification of marine mammals from Indian coasts. This was followed by a genetic study of Irrawaddy dolphin in Chilka Lake supported by Chilika Development Authority (Jayasankar et al., 2011). Two works on marine mammal taxonomy have been published from India (Jayasankar and Anoop, 2010; Vivekandandan et al., 2010). Standardized PCRbased methods for gender identification of species of marine mammals as well as in forensic identification of commercial products for checking illegal trade of the meat of endangered and protect species were developed as a result. Three important advancements from the molecular taxonomy approach during implementation of MoES-funded project, which could not have been possible with conventional approaches alone were (i) Correction of misidentification of species due to external body coloration differences between juveniles and adults [a specific case of Pantropical spotted dolphin], (ii) many beach-cast baleen whales in different stages of deterioration could be identified using DNA, and (iii) Sex [gender] of all samples were identified using PCR. Peer reviewed research papers and reviews on molecular identification and sex identification of marine mammals from Indian seas were published during 2007-2014 (Table 1).

Table 1. Particulars of marine mammal species from Indian seas which were identified using mtDNA markers

Species	Location	Number of individuals (n)	mtDNA gene	Reference
Tursiops aduncus	Vizhinjam, Kakinada & Chennai	5	Cytochrome b	Jayasankar <i>et al.</i> ,2008
Stenella longirostris	Kakinada & Chennai	12	Control region & Cytochrome b	_
Grampus griseus	Chennai	2	Control region & Cytochrome b	Jayasankar, 2014
Physeter macrocephalus	Chennai	2	Cytochrome b	_
Balaenoptera musculus	Mandapam	1	Control region & Cytochrome b	_
Dugong dugon	Mandapam	1	Control region & Cytochrome b	_
Stenella attenuata	Chennai	1	Control region & Cytochrome b	_
Delphinus capensis	Kakinada & Malpe	3	Cytochrome b	Jayasankar et al. 2008
Sousa chinensis (later described as S. plumbea)	Gangoli & Mangalore	2	Control region & Cytochrome b	_
Neophocaena phocaenoides	Gangoli, Malpe & Mangalore Thiruvananthapuram	7 1	Control region & Cytochrome b 16S rRNA & COI	Jayasankar <i>et al.</i> 2008 George <i>et al.</i> 2011
Balaenoptera edeni	Mandapam Thiruvananthapuram	1	Control region & Cytochrome b 16S rRNA & COI	Jayasankar <i>et al</i> . 2007 George <i>et al</i> . 2011
Orcaella brevirostris	Chilika Lake	11	Control region & Cytochrome b	Jayasankar <i>et al.</i> , 2011

#### Neophocaena phocaenoides

Finless porpoise (*N. phocaenoides*) is abundant in the west coast of India. This is the only representative of porpoises in Indian waters. Intraspecific genetic divergence is low when compared to some other dolphin species.



#### Stenella longirostris

Spinner dolphin (*S. longirostris*) is likely to be the most abundant dolphin in Indian waters as the molecular study conducted by the present author indicates. However, the species also exhibited maximum intraspecific genetic divergence. The taxonomy of *Stenella* is a matter of ongoing debate, and presence of multiple subspecies could further complicate the scenario.



#### **Tursiops aduncus**

Bottle nose dolphin (*T. aduncus*) was earlier mentioned as *T. truncatus* erroneously, and molecular study by the present author confirmed the species as *T. aduncus*. *T. truncatus* is larger than *T. aduncus* with a shorter beak. Certainly the species caught accidentally in gill nets is *T. aduncus*. However, among more oceanic species *T. truncatus* is likely to be present.



#### Delphinus capensis

Previously misidentified as *Delphinus delphis* from Indian waters, the long beaked common dolphin was re-described as *D. capensis* based on molecular study conducted by the present author. Intraspecific genetic divergence was found to be high in this species. Some confusion in absolute identity still remains, because some haplotype were closely similar to *D. tropicalis*, although *tropicalis* is treated as a sub-species by some experts, which means the Indian species of common dolphin is very likely to be *D. capensis tropicalis*.



#### New developments in molecular identification of marine mammals

Post 2014, the major technical advancement in molecular identification of marine mammals include application of DNA barcoding (COI, 16S rRNA), mass spectrometry (collagen peptide mass fingerprinting) and eDNA (droplet digital PCR). Next Gen Sequencing (NGS) has been more routinely applied to modern cetacean populations recovering full mitogenomes, genomic single nucleotide polymorphisms (SNPs), or even complete nuclear genomes to develop more nuanced models of their evolutionary systematics and population histories. Some of the current areas of molecular research on cetaceans globally are, DNA barcoding (Alfonsi et al., 2019), eDNA analysis (Baker et al., 2018), whole genome sequencing (Jia et al., 2019), mitogenomics (Cabrera et al., 2019) and molecular identification of market samples (Lee et al., 2019).

# Subspecies concept in marine mammal protection strategies

A species is a separately evolving lineage composed of a population or collection of populations. A subspecies is a population, or collection of populations, that appears to be a separately evolving lineage with discontinuities resulting from geography, ecological specialization, or other forces that restrict gene flow to the point that the population or collection of populations is diagnosably distinct (Taylor et al., 2017). Demographically Independent Population (DIP) means a sympatric group of individuals whose dynamics are more a consequence of births and deaths within the group (internal dynamics) than of immigration or émigration (external dynamics). DIP is an appropriate level of population structure for management objectives related to ecosystem function, like those of the U.S. Marine Mammal Protection Act (MMPA). Guidelines developed include reference to the need to provide information on the distributions of the taxa or taxon and on sample locations, descriptions of life history, and comments on choice of genetic markers and analytical methods. Such guidelines would improve consistency in the field of taxonomy. "Quantitative standards" are developed to illustrate the magnitude of differentiation that warrants subspecies classification. These standards would facilitate improvement of the quality and transparency of arguments advanced on behalf of taxonomic proposals and that they be viewed as "living standards" that can evolve with experience and as knowledge grows. Genetic metrics, such as Nei's estimate of net divergence (dA) and per cent diagnosability perform best to categorize cetacean populations, subspecies and species except in recently diverged species (Rosel *et al.*, 2017).

#### Way forward

Molecular identification attempts of cetaceans of Indian seas have clearly indicated the need for studying more number of species and individuals; phylogenetic relationships to understand the evolution of different species; and genetic variation *vis-à-vis* global geographic distribution of different species for their biodiversity conservation plans. In India we are continually monitoring stranding of cetaceans and dugong along beaches and landing centres. It is recommended that in addition to morphometric measurements and pictures of the specimens, a little quantity of skin tissue extracted from the specimens with minimum degree of deterioration is preserved in alcohol and molecular identification with standard methods is done for the credibility in identification of beach-cast or stranded marine mammals. Further, it is essential to venture into stock assessment of these gentle giants in our seas using non-invasive techniques like eDNA analysis. This is even more important in the context of conforming to global ocean conservation efforts like Marine Mammal Protection Act (MMPA).

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# Preliminary estimates of potential areas for seaweed farming along the Indian coast

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#### Abstract

India has enormous potential for seaweed mariculture; however, mass scale commercial farming of seaweeds is yet to take off successfully in the country. R&D efforts over the years have resulted in techno-scientific improvements in farming technologies such as floating rafts, net-tubes, long-lines, and cage based IMTA systems for seaweed culture. However, a few challenges remain, particular in identifying potential sites, its demarcation and developing suitable and sustainable spatial plans for seaweed farming on a country-wide, commercial perspective. In view of the emerging importance of seaweed mariculture and policy thrust by the Government of India, an all India preliminary site selection survey suitable for seaweed farming was conducted by ICAR-CMFRI along all maritime states of India. From this survey a total of 23,970 ha area were identified as potential seaweed farming along the Indian coast. In the present article, we present details of the suitable sites and its demarcation on a preliminary spatial map for facilitating the imminent expansion and effective adoption of seaweed farming in the country.

Keywords: GIS, mariculture, seaweed, site selection, spatial mapping

#### Introduction

Seaweeds are marine macroalgae which provide a variety of food products, phycocolloids (alginates, agars, and carrageenans), fodder and bio-fertilizers. Global seaweed production during 2018 was 32.4 million t (wet weight) with a first sale value estimation of 13.3 billion USD (FAO, 2020). Globally, seaweed farming has expanded rapidly due to its ever increasing demand and in India it is one of the best diversified-livelihood options for coastal fishers (Narayankumar and Krishnan, 2011). Various studies have been carried out on the potential of seaweed farming in India along with the available resource along various maritime states of India (Rao and Mantri 2006, Tandel *et al.*, 2016). These studies indicated that the major commercially important seaweed species in India are Gracilaria edulis, Gelidiella acerosa and Kappaphycus alvarezii in red algae and Sargassum wightii, Turbinaria conoides and Cystoseira spp. in brown algae. Besides some of the green algae like Ulva lactuca, Enteromorpha sp., Caulerpa spp. which can be used for human consumption and can be part of the regular diet for nutritional security. However, the pace of seaweed farming in India has been constrained due to inadequate marine spatial plans which needs a systematic site selection process. In this context, ICAR-Central Marine Fisheries Research Institute (CMFRI) initiated a preliminary survey all along the coastal regions of the country for identifying potential seaweed farming areas. Initial assessments on the potential areas for seaweed farming were conducted through informal surveys during field visits by scientific and technical personnel along with the information collected from local fishers through personal interactions.

Site selection plays an important role in the success of any sustained commercial farming activity. It significantly influences the economic returns and viability of the farming system. In the same manner seaweed farming also needs best suitable farming sites for successful operation. Although Divu *et al.*, (2020) developed a novel GIS based site suitability model for mariculture in territorial waters of the country, the candidates for their model were marine finfish and shellfish species and the model could not cover seaweeds. Thus this preliminary survey was conducted as a first step towards getting baseline data for future development of spatial models and spatial plans for seaweed mariculture in India. Site suitability was worked out for all maritime states along the Indian coast. The methodology and criteria for the site suitability are mentioned below.

## Criteria used for identifying the potential seaweed farming sites:

- Nearshore area within 1000 m distance from the lowest low tide line
- Intertidal and sub-tidal zones with rocky or sandy bottom
- Previous existence of seaweed farming activity (if any along the coast)
- Seaweed collection from natural seaweed beds (if existing)
- Sheltered area with adequate current and tidal exchange
- Area with moderate wave action
- Area free from silt deposits
- Area away from freshwater runoff and domestic or agro-industrial effluents discharge
- Area away from fishing harbor/landing centre
- Non-hindrance for existing fishing and other allied activities
- Optimum basic water quality parameters: Salinity (28-38 ppt), Sea Surface Temperature (26-31°C), pH (6.5-8.5) and Transparency (2-6 m).

Considering the above-mentioned criteria, preliminary identification of the potential sites for seaweed farming along the Indian coast was made. The potential area and

State	No. of locations identified	Preliminary demarcation of potential sites (in ha)
Gujarat	9	10316
Diu	5	700
Maharashtra	12	2724
Goa	4	120
Karnataka	14	1579
Kerala	7	80
Lakshadweep Islands	11	213
Total West Coast	62	15,732
Tamil Nadu	187	5048
Andhra Pradesh	49	1215
Odisha	14	1525
West Bengal	5	450
Total East Coast	255	8238
Total (All India)	317	23,970

Table 1. Potential seaweed farming sites along Indian coast

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production potential will vary from site to site depending on the local climatic conditions and number of farming cycles in a year.

## State-wise potential area available for seaweed farming

The geo-morphology and demography of India's coastline is diverse and distinct. Each maritime state

has its individual advantages and disadvantages with respect to seaweed farming. Since this study was a preliminary assessment, broader arrays of biological and environmental parameters have been taken as site selection criteria. The information is represented as the name of the village/site, name of the district, its location with latitude and longitude and approximate area available for seaweed farming in hectare (ha).

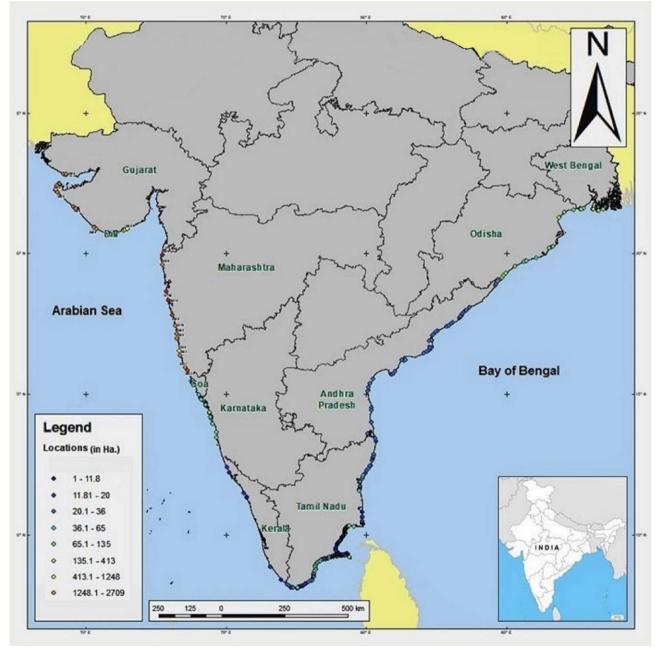


Fig 1. Potential seaweed farming locations in various maritime states of India

Table 2. Potential areas for seaweed farming in Gujarat

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Kutchh District		
Mandvi	22°50′12.6″N, 69°12′17.4″E	1500
Total available area for Kutchh District		1500
Dwarka District		
Dwarka	22°15′33.1″N, 68°55′26.4″E	2000
Okha	22°29′13.7″N, 69° 2′39.4″E	2000
Total available area for Dwarka District		4000
Amreli District		
Jafrabad	20°50′19.8″N, 71°20′38.1″E	616
Total available area for Amreli District		616
Gir-Somnath District		
Madhwad-Site 1	20°41′31.5″N, 70°50′44.2″E	300
Madhwad-Site 2	20°41′59.1″N, 70°51′43.2″E	200
Madhwad-Site 3	20°41′10.7″N, 70°50′16.0″E	200
Veraval	20°55′54.6″N, 70°18′53.7″E	2000
Total available area for Gir-Somnath District		2700
Porbundar District		
Porbandar	21°38′53.1″N, 69°34′4.8″E	1500
Total available area for Porbundar District		1500
Total available area in Gujarat		10316

#### Table 3. Potential areas for seaweed farming in in Diu (UT)

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Simar	20°44′46″N,71° 5′12 "E	200
Navbundar-Site 1	20°43′35″N, 71° 2′24″E	50
Navbundar-Site 2	20°43′49″N, 71° 3′55″E	50
Chakrathirth coast	20°42′4″N, 70°56′21 "E	300
Vanakbara coast	20°41′41″N, 70°53′39″E	100
Total Area available in Diu		700

Table 4. Potential areas for seaweed farming in in Maharashtra

Name of the location	GPS Coordinates (D.M.S)	Total available are (in ha) (approx.)
Palghar District		
Dhanu	19° 57′ 36′′ N, 72° 43′ 48′′ E	10267
Kelva	19° 36′ 36′′ N, 72° 43′ 48′′ E	2709
Total available area for Palghar District		12976
Raigad District		
Alibaug	18° 38′ 60″ N, 72° 51′ 36″ E	12198
Murud	18° 19′ 48′′ N, 72° 57′ 36′′ E	9415
Total available area for Raigad District		21613
Ratnagiri District		
Harnai	17° 48′ 36′′ N, 73° 5′ 24′′ E	5200
Guhaghar	17° 25′ 48′′ N, 73° 11′ 24′′ E	2116
Ganpatiphule	17° 8′ 24′′ N, 73° 15′ 36′′ E	1583

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Total available area for Ratnagiri District		8899
Sindhudurg District		
Vijaydurg	16° 33′ 0′′ N, 73° 19′ 48′′ E	1248
Devgad	16° 22′ 12′′ N, 73° 22′ 12′′ E	1663
Achara	16° 59′ 24′′ N, 73° 26′ 24′′ E	2200
Shriramwadi	15° 56′ 24′′ N, 73° 32′ 60′′ E	2347
Vengurla	15° 50′ 24′′ N, 73° 37′ 48′′ E	3533
Total available area for Sindhudurg District		10991
Total area available in Maharashtra		54479*
Area accounted for present purpose (5%)		2724

\*Since it is preliminary assessment only 5% of the suitability taken in to account for immediate support

Table 5. Potential areas for seaweed farming in in Goa

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
North Goa District		
Siridoa	15°25′49′′ N, 73°52′ 01′′ E	7.5
	15°25′52′′ N, 73°52′ 04′′ E	
Caranzalem	15° 27′36′′ N, 73° 45′ 52′′ E	63
	15° 28'40'' N, 73° 48' 22'' E	
Total available area for North Goa District		70.5
South Goa District		
Baina	15° 23′43′′ N, 73° 48′ 13′′ E	4
	15° 23′37′′ N, 73° 48′ 19′′ E	
Talpona	14° 58′24′′ N, 74° 02′ 35′′ E	45
	14° 58′56′′ N, 74° 02′ 20′′ E	
Total available area for South Goa District		49
Total area available in Goa		120

#### Table 6. Potential areas for seaweed farming in Karnataka

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Jttara Kannada District		
Dhandebag-Kangiguda Island, Karwar	14° 53′19′′ N, 74° 05′ 59′′ E	101
Baval-Kanga Island, Karwar	14° 51′56′′ N, 74° 06′ 29′′ E	11
Harwada, Ankola	14° 42′50′′ N, 74° 15′ 49′′ E	72
Belikeri, Ankola	14° 42′ 14″ N, 74° 15′ 54″ E	135
Gabit Keni, Ankola	14° 39′ 46′′ N, 74° 16′ 41′′ E	7
Belambar, Ankola	14° 38′ 52′′ N, 74° 16′ 38′′ E	244
Haldipur-Horbhag, Honnavar	14° 18′ 44′′ N, 74° 24′ 53′′ E	413
Manki 1, Honnavar	14° 11′ 27′′ N, 74° 28′ 04′′ E	50
Manki 2, Honnavar	14° 8′ 29′′ N, 74° 28′ 43′′ E	94
Navayatkeri, Murudeshwara (North)	14°11′85″N, 74 <sup>0</sup> 27′40″ E	52
Huddi Point South Bhatkal-Shiroor North)	14º56′85"N, 74º32′98"E	100
otal available area for Uttara Kannada District		1279

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Udupi District		
Navunda South	14°42′30″N 74°38′44″E	50
Kundapur	13°39′.62″N 74°39′25″E	120
Hoode	13°27′42″N 74°40′42″E	130
Total available area for Udupi District		300
Total Area available in Karnataka		1579
Table 7. Potential areas for seaweed farming in in Kerala		
Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)
Thiruvananthapuram District		
Vizhinjam	8°23′1.24″ N, 76°57′36.67″E	10
Total available area for Thiruvananthapuram District		10
Kollam District		
Thirumallavaram	8°54′42″N, 76°38′ 21″E.	20

Total available area for Kollam District		20
Kozhikode District		
Elathur	11°20′07.03″ N, 75° 44′35″ E	1
Puthiyappa	11°19′18.17″ N, 75°44′24.65″ E	7
Thikkodi	11°28′ 46.1″N, 75°37′ 28.8″E	20
Total available area for Kozhikode District		28
Kasargod District		
Padanna	12° 12′20.52″ N; 75° 07′22.22″ E	5
Bekal	12°23'43.8"N; 75°02'78"E	17
Total available area for Kasargod District		22
Total area available in Kerala		80

Table 8. Potential areas for seaweed farming in Tamil Nadu

Name of the location	GPS coordinates (D.M.S)	Total available area (ha) (approx.)
Ramanathapuram District (Palk Bay)		
Dhanushkodi (Pachapatti)	9°11′41.7″N, 79°24′18.9″E	90
Sangumal	9°17′40.1″N, 79°19′36.3″E	25
Olaikuda	9°19′01.2″N, 79°19′54.9″E	34
Mangadu	9°19'39.0″N, 79°18'55.1″E	22
Sambai	9°19′41.7″N, 79°18′46.0″E	30
Vadakadu	9°19′22.2″N, 79°17′59.8″E	30
Pillaikulam	9°19′15.3″N, 79°17′34.5″E	26
Ariyankundu	9°17′52.6″N, 79°16′19.1″E	23
Villoondi	9°17′33.9″N, 79°15′41.9″E	26
Manthoppu	9°17′30.4″N, 79°15′14.4″E	14
Victoria Nagar	9°17′32.2″N, 79°14′42.3″E	9.5
Naalupanai	9°17′32.3″N, 79°14′22.8″E	15

me of the location	GPS coordinates (D.M.S)	Total available are (ha) (approx.)	
Akkalmadam	9°17′31.7″N, 79°13′56.6″E	20	
Pamban	9°17′29.1″N, 79°13′13.0″E	8	
Thonithurai	9°17′02.0″N, 79°10′45.7″E	14	
Meenavar colony	9°17′04.2″N, 79°10′26.6″E	6	
T.Nagar	9°17′29.0″N, 79°08′40.9″E	15	
Munaikadu	9°17′16.1″N, 79°07′59.8″E	40	
Umayalpuram	9°17′15.5″N, 79°07′31.7″E	38	
Vedalai	9°17′20.4″N, 79°06′18.0″E	24	
Pillaimadam	9°17′41.9″N, 79°05′07.2″E	22	
Pirappanvalasai	9°18′21.0″N, 79°03′15.3″E	16	
Irumeni	9°19′21.4″N, 79°01′43.8″E	16	
Uchipuli	9°19′59.3″N, 79°00′55.4″E	20	
Attrangarai	9°21′03.7″N, 78°59′35.7"E	15.3	
Alakankulam	9°21′51.8″N, 78°58′43.8″E	15.9	
Panaikulam	9°22′40.7″N, 78°57′57.4″E	16	
Puduvalasai	9°23′46.4″N, 78°56′55.9"E	19	
Athiyuthu (Iraniyanvalasai)	9°24′27.2″N, 78°56′20.5"E	15	
Palanivalasai	9°25′10.9″N, 78°55′46.2″E	9	
Mudiveeranpattinam	9°26′46.1″N, 78°54′46.7″E	27	
Devipattinam	9°29'17.4″N, 78°53'53.1″E	2	
Thiruppalaikudi	9°32′12.1″N, 78°55′07.4″E	8	
Karankadu	9°38′46.9″N, 78°57′57.0″E	8.5	
Mullimunai	9°39′19.7″N, 78°58′13.5″E	9	
Puthupattinam (K.K. Pattinam)	9°40′33.3″N, 78°58′29.9″E	12	
Veerasangili Madam	9°41′13.6″N, 78°58′46.9″E	23	
Soliyakudi	9°42′48.3″N, 78°59′56.9″E	15	
Nambuthalai	9°43′44.1″N, 79°00′47.3″E	7.5	
Thondi	9°45′02.5″N, 79°01′42.3″E	10.5	
M.R.Pattinam	9°45′42.6″N, 79°02′11.4″E	12	
P.V.Pattinam	9°45′59.7″N, 79°02′33.5″E	9.8	
Narenthal	9°46′08.8″N, 79°03′02.8″E	13	
Vattanam	9°47′09.5″N, 79°03′53.6″E	20	
Dhamothirapattinam	9°47′38.2″N, 79°04′13.8″E	14	
Pasipattinam	9°48′16.0″N, 79°04′45.4″E	12	
Theerthandatnam	9°49′32.9″N, 79°05′22.8″E	8	
S.P.Pattinam	9°50′07.7″N, 79°06′09.1″E	15	
al available area for Ramanathapuram District (Palk Bay)		900	
manathapuram District (Gulf of Mannar)			
Kunthukal	9°15′48.5″N, 79°13′16.0″E	20	
Mandapam	9°16′34.1″N, 79°08′45.2″E	18	

me of the location	GPS coordinates (D.M.S)	Total available are (ha) (approx.)	
Vedalai	9°15′37.4″N, 79°05′29.7″E	30	
Seeniappa Dharga	9°15′40.0″N, 79°04′03.8″E	24	
Nochioorani	9°16′00.8″N, 79°02′05.8″E	19	
Manankudi	9°16′16.8″N, 79°00′25.1″E	16 25	
Pudumadam	9°16′24.4″N, 78°59′03.4″E		
Valangapuri	9°16′22.5″N, 78°58′01.0″E	12.5	
Vellarioodai	9°16′20.3″N, 78°57′24.7″E	15	
Thalai Thoppu	9°16′13.8″N, 78°56′42.0″E	20	
Inthira Nagar	9°15′45.4″N, 78°55′15.1″E	12	
Munthal (Periyapattinam)	9°15′08.1″N, 78°54′41.6″E	13	
Pudhukudiyiruppu (Periyapattinam)	9°15′08.5″N, 78°53′47.7″E	10	
Thoppuvalasai	9°15′16.8″N, 78°53′16.4″E	15	
Velayuthapuram	9°15′20.7″N, 78°52′55.6″E	13.5	
Kalimankundu	9°15′14.5″N, 78°51′58.5″E	10	
Sethukarai	9°14′54.4″N, 78°50′41.4″E	8.5	
Kanjirangudi (Pakkirappa Dharga)	9°14′33.4″N, 78°49′42.7″E	14	
Sengalaneerodai	9°14′13.3″N, 78°48′44.8″E	25	
Keelakarai	9°13′26.5″N, 78°46′32.8″E	22 25	
Bharathinagar	9°12′59.6″N, 78°45′26.6″E		
Mangaleswari Nagar	9°12′41.3″N, 78°44′05.2″E	28	
Earanthurai	9°12′24.6″N, 78°43′31.0″E	26	
Erwadi	9°11′59.8″N, 78°43′15.8″E	18.5	
Sadaimuniyanvalasai	9°11′27.8N, 78°42′37.3″E	16	
P.M. Valasai	9°11′35.9″N, 78°41′52.8″E	36	
Adancheri	9°11′39.1″N, 78°39′48.8″E	28	
Valinokkam	9°09′13.6″N, 78°37′41.8″E	88	
Keelamundhal	9°08′26.6″N, 78°35′26.4″E	30	
Melamundhal	9°07′59.7″N, 78°34′12.6″E	31	
Mariyur	9°08′12.4″N, 78°32′31.0″E	34	
Oppilan	9°08′04.3″N, 78°30′41.9″E	29.5	
Mookaiyur	9°07′39.0″N, 78°28′38.6″E	30	
Naripaiyur	9°07′06.7″N, 78°25′51.8″E	24	
Kannirajapuram	9°06′19.3″N, 78°24′08.8″E	28.5	
Rochma Nagar	9°05′47.3″N, 78°23′23.5″E	35	
al available area for Ramanathapuram District (Gulf of Mannar)		850	
al available area for Ramanathapuram District (Palk Bay & Gulf of Manr	nar)	1750	
dukottai District (Palk Bay)			
Muthukuda	9°52′30.8″N, 79°07′07.5″E	7.2	
Arasanagaripattinam	9°53′37"N 79°07′38″E	35	
Mimisal	9°54'42″N, 79°08'50″E	22	

Name of the location	GPS coordinates (D.M.S)	Total available area (ha) (approx.)	
Gopalapattinam	9°55′26″N, 79°09′10″E	15	
Palakkudi	9°56′37″N, 79°10′06″E	18.5	
Kallivayal (Muthanenthal)	9°57′12″N 79°10′37″E	17.6	
Jagathapattinam	9°57′58″N, 79°11′24″E	10.4	
Kottaipattinam	9°58′40″N, 79°12′02″E	15.5	
Odavimadam	9°59′15″N, 79°12′30″E	16.6	
Pudukkudi	10°00′03″N, 79°13′14″E	14	
Aathipattinam	10°00'26"N, 79°13'36"E	12.4	
Ammapattinam	10°00′54.3″N, 79°13′59.8″E	14	
Avudaiyarpattinam	10°01′13″N, 79°14′22″E	19	
Sangupattinam (Rajathoppu)	10°01′51.7″N, 79°15′05.4″E	5.5	
Kodiyakarai (Manamelkudi)	10°02′05″N, 79°15′30″E	23	
Muthurajapuram (Manamelkudi)	10°02′23.7″N, 79°15′49.6″E	22	
Seetharamanpattinam	10°04′29″N, 79°14′11″E	10	
Krishnajipattinam	10°05′48″N, 79°13′38″E	12	
P.R.Pattinam	10°06′08.3″N, 79°13′39.7″E	10.3	
otal available area for Pudukottai District		300	
nanjavur District (Palk Bay)			
Ganeshapuram	10°08′14.0″N, 79°13′43.4″E	7.1	
Somanathapattinam	10°09′30.8″N, 79°14′25.7″E	7.5	
Mandhiripattinam	10°10′18.4″N, 79°14′24.6″E	9	
Senthalaipattinam	10°11′12″N, 79°14′55″E	14	
Adaikathevan	10°12′00.3″N, 79°15′56.3″E	8.5	
Karankuda	10°14′17.0″N, 79°16′18.6″E	9.2	
Sethubavachathiram	10°15′08″N, 79°17′11″E	12	
Pillayarthidal	10°15′26″N, 79°17′30″E	17	
Manora	10°15′55″N, 79°18′9.999″E	10.5	
Chinnamanai	10°16′08″N, 79°18′38″E	2.2	
Mallipattinam	10°16′50″N, 79°19′27″E	20	
Pudhupattinam	10°17′11.2″N, 79°20′15.0″E	26	
Kollukadu	10°17′30.3″N, 79°21′46.8″E	34	
Athiramapattinam	10°18′59″N, 79°23′46″E	73	
otal available area for Thanjavur District		250	
niruvarur District (Palk Bay)			
Thondiyakadu	10°23′23″N, 79°34′46″E	100	
available area for Thiruvarur District		100	
agapattinam District (Palk Bay)			
Maniyantheevu	10°21′37.4″N, 79°52′27.8″E	28	
Arcottuthurai	10°23′53″N, 79°52′09″E	40	
Periyakuthagai	10°24′50″N, 79°52′01″E	54	
Pushpavanam	10°27′22″N, 79°51′50″E	74	

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Name of the location	GPS coordinates (D.M.S)	Total available area (ha) (approx.)		
Naluvethapathi	10°29′07″N, 79°51′46″E	20		
Vizhunthamavadi	10°35′57″N, 79°51′23″E	18		
Kameshwaram	10°37′27″N, 79°51′14″E	13		
Sammanthan Pettai	10°47′31″N, 79°51′03″E	3		
otal available area for Nagapattinam District		250		
uticorin District (Gulf of Mannar)				
Vembar	9°05′00.10″N, 78°22′30.02″E	80		
Periyasamypuram	9°02′58.85″N, 78°20′09.03″E	50		
Keelavaippar	9°00'02.62″N, 78°15'52.61″E	60		
Sippikulam	8°58′57.09″N, 78°14′13.18″E	75		
Pattinamaruthur	8°56′17.00″N, 78°11′39.86″E	80		
Tharavaikkulam	8°53′34.34″N, 78°10′47.43″E	70		
Vellapatti	8°51′48.48″N, 78°10′10.43″E	60		
Mottagopuram	8°50'44.02"N, 78°10'02.60"E	40		
Tuticorin Harbour Point	8°46′33.72″N, 78°12′07.72″E	80		
Mullakaadu	8°44′25.90″N, 78°10′10.58″E	90		
Palayakayal	8°41′31.85″N, 78°08′20.35″E	50		
Punnakayal	8°36′41.49″N, 78°07′48.75″E	25		
Kayalpattinam	8°33′58.69″N, 78°08′04.47″E	80		
Veerapandiyapattinam	8°30′51.40″N, 78°07′28.11″E	60		
Amali Nagar	8°29'25.15"N, 78°07'38.41"E	30		
Alanthalai	8°25'47.12"N, 78°04'25.06"E	80		
Kulasekarapattinam	8°23′39.30″N, 78° 3′30.58″E	40		
Manapadu	8°22′31.01″N, 78° 3′51.76″E	65		
Periyathalai	8° 21′34.63″E, 78° 1′41.60″E	35		
otal available area for Tuticorin district		1150		
irunelveli District				
Periyathalai	8°17′49.28″N, 77°55′40.18″E	35		
Kootapanai	8°15′44.47″N, 77°51′51.38″E	15		
Kooduthalai	8°14'59.18"N, 77°49'31.91"E	15		
Uvari	8°13′26.16″N, 77°47′14.79″E	20		
Idinthakarai	8°11′05.33″N, 77°45′27.38″E	15		
Kuthenkuli	8°09'47.27"N, 77°41'21.49"E	15		
Perumanal	8°09′28.75″N, 77°38′49.18″E	15		
Kootapuli	8°08′46.97″N, 77°36′24.73″E	10		
Thomaiyarpuram	8°08′19.50″N, 77°35′2.19″E	10		
otal available area for Tirunelveli District		150		
anyakumari District				
Thengapattinam	8°14′11.40″N, 77°10′14.61″E 30			
Colachel	8°10′20.66″N, 77°15′12.65″E 30			
Kadiapattinam	8° 7′53.28″N, 77°18′13.81″E	30		

Name of the location	GPS coordinates (D.M.S)	Total available area (ha) (approx.)	
Muttom	8° 7′15.59″N, 77°19′11.22″E	70	
Pillaithoppu	8°07′29.08″N, 77°20′01.97″E	20	
Periyakaadu	8°06′31.93″N, 77°23′38.74″E	30	
Kovalam	8°04′50.20″N, 77°31′37.60″E	20	
Kanyakumari	8°05′07.69″N, 77°33′11.41″E	40	
Chinnamuttom	8°06′05.23″N, 77°33′29.80″E	30	
Arokiapuram	8°06′20.15″N, 77°33′31.28″E	50	
otal available area for Kanyakumari District		350	
uddalore District			
Sonankuppam	11° 43′ 25″ N, 79° 46′ 59″ E	20	
Singarathope	11° 43′ 11″ N, 79° 46′ 56″ E	35	
Rajapettai	11° 40′ 57″ N, 79° 46′ 24″ E	50	
Chithiraipettai	11° 38′ 15″ N, 79° 45′ 49″ E	25	
Thamanam pettai	11° 37′ 10″ N, 79° 45′ 38″ E	50	
Annappan pettai	11° 35′ 11″ N, 79° 45′ 31″ E	35	
Kumarapettai	11° 34′ 20″ N, 79° 45′ 30″ E	25	
Samiyarpettai	11° 32′ 59″ N, 79° 45′ 38″ E	50	
otal available area for Cuddalore District		290	
/illupram District			
Bommaya palayam	11° 59′ 24″ N, 79° 51′ 05″ E	25	
Koonimedu	12° 04′ 44″ N, 79° 53′ 43″ E	50	
Anumandai	12° 07′ 29″ N, 79° 55′ 25″ E	45	
Ekkiyarkuppam	12° 10′ 55″ N, 79° 57′ 44″ E	20	
otal available area for Villupuram District		140	
Chengalpattu District			
Edaikazhinadu	12° 17′ 37″ N, 80° 01′ 43″ E	25	
Paramankeni	12° 20′ 45″ N, 80° 04′ 01″ E	25	
Kadalur Chinna kuppam	12° 26′ 54″ N, 80° 08′ 43″ E	25	
Kadalur Periya kuppam	12° 26′ 31″ N, 80° 08′ 18″ E	33	
Devaneri	12° 39′ 00″ N, 80° 12′ 31″ E	35	
Nemmeli	12° 42′ 49″ N, 80° 13′ 55″ E	30	
Semencheri	12° 44′ 25″ N, 80° 14′ 27″ E	20	
Kovalam	12° 47′ 26″ N, 80° 15′ 10″ E	50	
Kanathur	12° 51′ 58″ N, 80° 15′ 02″ E	30	
otal available area for Chengalpattu District		273	
hiruvallur District			
Kalanji	13° 19′ 53″ N, 80° 20′ 36″ E	20	
Pulicut	13° 25′ 14″ N, 80° 19′ 46″ E	25	
otal available area for Thiruvallur District		45	
otal Area available in Tamil Nadu		5048	

Table 9. Potential Areas for Seaweed Farming in Andhra Pradesh

Name of the location	GPS Co-ordinates (D.D)	Total available area (in ha) (approx.)	
Visakhapatnam District			
RK Beach	17.715 N, 83.325 E	40	
VUDA Park	17.722 N,83.340 E	10	
Tenneti Park	17.747 N,83.350 E	50	
Thotlakonda	17.772 N,83.378 E	25	
Bhimli	17.892 N,83.455 E	25	
Thimmapuram	17.813 N,83.411 E	50	
Mangamaripeta	17.838 N,83.411 E	50	
Yendada	17.769 N,83.372 E	25	
Muthyalammapalem	17.535 N,83.090 E	25	
Pudimadaka	17.491 N,83.004 E	50	
Bangarammapalem	17.413 N, 82.859 E	25	
Rambilli	17.447 N, 82.933 E	25	
otal available area for Visakhapatnam District		400	
/ijayanagaram District			
Mukkam	17.989 N, 83.560 E	35	
Kancheru	17.964 N, 83.544 E	30	
Bhogapuram	17.978 N, 83.554 E	40	
Musalayya palem	17.764 N, 83.364 E	35	
Neelagaddapeta	18.087 N, 83.688 E	25	
otal available area for Vijayanagaram District		165	
irikakulam District			
Baruva-Kothuru	18.878 N, 84.593 E	50	
Sompeta	18.918 N, 84.630 E	25	
otal available area for Srikakulam District		75	
ast Godavari District			
Uppada	17.078 N, 82.338 E	25	
Konapapapeta	17.132 N, 82.395 E	35	
Pampodipeta	17.243 N, 82.533 E	30	
Cholangi	16.898 N, 82.244 E	25	
Mulapeta	17.104 N, 82.365 E	35	
Danaiahpeta	17.215 N, 82.493 E	50	
Narsipeta	17.212 N, 82.489 E	25	
Neelarevu and Pandi	16.539 N, 82.223 E	25	
otal available area for East Godavari District		250	
Vest Godavari District			
Vemuladeevi	16.195 N, 81.355 E	50	
Perupalem	16.202 N, 81.355 E	50	
otal available area for West Godavari District		100	
Krishna District			
Urlagondadibba	16.205 N, 81.255 E	50	
Chinnagollapalem	16.213 N, 81.405 E	25	

Name of the location	GPS Co-ordinates (D.D)	Total available area (in ha) (approx.) 30 105	
Sorlagondi	15.824 N, 80.988 E		
Total available area for Krishna District			
Prakasam District			
Rajupalem	15.137 N, 80.061 E	25	
Ethamukkala	15.372 N, 80.125 E	25	
Ullapalem	15.242 N, 80.085 E	25	
Total available area for Prakasam District		75	
SPSR Nellore District			
Mypadu	14.506 N, 80.179 E	20	
Kothapallipalem	14.442 N, 80.175 E	25	
Total available area in SPSR Nellore District		45	
Total Area Available in Andhra Pradesh		1215	

Table 10. Potential Areas for Seaweed Farming in Odisha

Name of the location	GPS Coordinate (D.D)	Total available area (in ha) (approx.)	
Puri District			
Chilka lake Arakuda (Near Bar mouth area)	19.7329°N, 85.67939°E	50	
Satpada	19.70856°N, 85.62587°E	125	
Ramchandi Muhanan near Chandrabhaga	19.854580°N, 86.059211°E	50	
Baliharichandi area	19.74802 N, 85.69988 E	50	
Total available area for Puri District		275	
Ganjam District			
Puruna bandha area	19.2899° N, 84.98094° E	150	
Ramayapatnam	19.15088°N, 84.83727° E	150 200	
Kalijai area	19.53661° N, 85.30235° E		
Gopalpur Open sea	19.22097° N, 84.88213° E	100	
Total available area for Ganjam District		600	
Baleswar District			
Balaramgadi to Mahi sahi area	21.47339°N, 87.0557°E	100	
Balarampur Panchubisha to Januka	21.27523°N, 86.86788°E	150	
Kirtania to Talasari	21.56294°N, 87.388°E	100	
Total available area for Baleswar District		350	
Jagatsingpur District			
Jatadhari Muhana Gadakujanga	20.215°N, 86.61137°E	150	
Sea Near Neheru Banglow	20.24755° N, 86.61137° E	50	
Gada Harishpur	20.18932°N, 86.52473°E	100	
Total available area for Jagatsingpur District		300	
Total Area Available in Odisha		1525	

Table 11. Potential Areas for Seaweed Farming in West Bengal

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)	
South 24 Praganas District			
Fraserhanj (Bakkhali)	21° 31′ 41″N, 88° 15′ 52″ E	100	
Sagar Island Systems	21° 35′ 16″N, 88° 04′ 18″E	125	
Sundarban Dhanchi Forest	21° 34'42"N, 88° 25'45"E	95	
Total available area for South 24 Parganas District		320	
Purba Medinipur District			
Mandarmani	21° 36′ 14″N, 87° 43′ 29″ E	70	
Shankarpur	21° 35′ 33″N, 87° 37′ 12″E	60	
Total available area for Purba Medinipur District		130	
Total Area Available in West Bengal		450	

Table 12. Potential area for seaweed farming in Lakshadweep

Name of the location	GPS Coordinates (D.M.S)	Total available area (in ha) (approx.)* 17.5	
Agatti	10° 51′N, 72° 11′E		
Amini	11°07′N, 72°43′E	1.5	
Androth	10°48′N, 73°40′E	0.5	
Bitra	11°35′N, 72°11′E	45.6	
Bangaram	10 ° 56′N, 72° 17′E	46.3	
Chetlath	11 ° 41′N, 72° 43′E	1.6	
Kiltan	11 ° 29′N, 72° 59′E	1.8	
Kadmath	11 ° 12′N, 72° 45′E	37.5	
Kalpeni	10°04′N, 73°37′E	25.6	
Kavaratti	10°33'N, 72°38'E	5.0	
Minicoy	8°70′N, 73°03′E	30.6	
otal Area Available in Lakshadweep Islands		213.5	

\* Atoll-wise (all inhabited atolls) area of lagoon and one percentage (area suitable for farming)

## Actions to be undertaken before implementing seaweed farming

The identified areas must be precisely modelled using GIS based studies by considering the physico-chemical and biological parameters for the identified locations prior to the mass scale implementation of this farming activity. Necessary permission may be obtained in the Biosphere Reserves/Marine Protected Areas including marine national parks and sanctuaries if any, prior to seaweed farming implementation. Local community consensus through stakeholder consultations has to be obtained prior to implementation of seaweed farming activities. Wherever possible, seaweed farming area needs to be demarcated to avoid sectoral and spatial conflict with other livelihood activities. Pilot scale farming can be undertaken to study the suitability of seaweed species and farming methods in each of the identified sites before large scale implementation of the programme. Impact assessment studies of seaweed farming (e.g. corals, seagrass, etc.) must be carried out. Infrastructure for drying and storing of seaweeds and marketing channels also need to be created for success of seaweed farming in the country.

Expansion of seaweed farming as an additional livelihood option in the Indian coastal region will pave the way for socioeconomic upliftment of coastal fishers/farmers. Further it will be helpful for mitigating the negative effects of climate change along with many other natural benefits. Owing to the importance of seaweed, the Government of India is promoting seaweed farming and its related activities through the recently launched flagship programme *Pradhan Mantri Matsya Sampada Yojana* (PMMSY) by providing financial, marketing and logistical support. Thus this is the ideal moment to take seaweed farming forward in the country.

#### **Recommendations and Way forward**

The current study is a preliminary assessment only. In order to explore suitable sites for seaweed culture in detail, it is necessary that the available sea space be modelled by using advanced computational tools like GIS. Site suitability indexes need to be developed for seaweed farming systems. Along with this, species-specific analysis must be developed for further sustainable planning for expansion of this activity in a commercial manner.

Comprehensive planning for seaweed farming in the territorial waters needs to be carried out. This must be performed by considering the opinions of wide range of stakeholders along with the existing coastal communities' acceptance of this activity through technology demonstration and validation. Unexplored sheltered Island waters need to be explored for seaweed farming by considering all potential impacts over its specific existing sensitive ecosystems. Lagoons, the shallow and sheltered area in the atoll islands of Lakshadweep is ideal for seaweed farming. An approximate area of 213.4 ha has been preliminarily identified at Lakshadweep waters (in all the 11 inhabited Islands) and studies are progressing at Andaman and Nicobar Islands. Due to geographic and ocean climate advantages it is suggested that 10% of lagoon areas of the islands can be used for seaweed farming. In the island ecosystems, we recommend farming of native seaweed species only.

Development of analytical tools for spatial management is the need of the hour. Therefore, future research can focus on development of spatial management tools which could provide decision makers with a sciencebased objective tool to harness the ocean sustainably. As the current study is only a preliminary approach for obtaining site suitability for seaweed farming by taking into consideration suitable water guality parameters for culture, there are chances that many sites which may be suitable for culture might not have been included in this assessment. The current study can also be considered as a guide for further studies in these lines. The site suitability studies for seaweed farming needs a detailed and comprehensive analysis including experimental farming, consultation of stakeholders and coastal communities involved in the various seaweed farming activities, considering the constraints such as marine protected areas, marine national parks, impact assessment studies on other fauna and flora, feeding and breeding grounds of some specific region for protected marine species such as Olive Ridley turtles along Odisha coast and also the natural disasters. As the coastal conditions along various maritime states are not uniform, it is very important that the assessment needs to proceed by taking into consideration all region-specific aspects while developing the final model for seaweed farming along the Indian coast.

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ICAR-CMFRI | Marine Fisheries Information Service Technical & Extension Series No. 246, 2020

### Development of a multivalent vibriosis vaccine and its application in sea cage farming of cobia

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#### Abstract

The loss due to diseases in the aquaculture sector is estimated to be around 10-15% of the production cost and it can be substantially reduced if due attention is given to scientific health management. Most of the antibiotics are banned in aquaculture, so the alternative to maintain fish health is based on the supplementation of probiotics, immunostimulants and administration of vaccines. The main objective of developing a multivalent vaccine against vibriosis is to prevent the seasonal epizootics in cultured cobia *Rachycentron canadum*. The whole cell inactivated multivalent vibrio vaccine against vibriosis was developed, standardized and evaluated in cobia fingerlings. The field application of the vaccine was studied in sea cages deployed for growout and broodstock rearing of cobia. The immune responses were evaluated by challenge studies in fingerlings and serum antibody titre in cobia growout and broodstock cages using ELISA. The regular epizootics observed in cage cultured cobia, every year during the months of July to September (pre-monsoon season) was prevented by timely vaccination and proper sea cage farming management.

Keywords: Cobia, vibriosis, multivalent vaccine, cage farming

#### Introduction

Breeding, seed production and farming technologies for two marine finfishes namely cobia (Gopakumar *et al.*,2011) and silver pompano were developed by ICAR-Central Marine Fisheries Research Institute (CMFRI). The techno economic viability of farming these fishes in sea cages and ponds reached the fishermen and fish farmers through different training, awareness and frontline/participatory demonstration programmes organised by the Mandapam Regional Centre of ICAR -CMFRI. In view of the declining catches, sea cage farming can enhance the marine fish production and help to increase the income of the fishers. Currently, the institute is extending technology support to interested fishermen and entrepreneurs for cage farming and hatchery technology required for seed production of high value marine finfishes, in several maritime states such as Tamil Nadu, Karnataka, Andhra Pradesh, Kerala, Goa, Maharashtra, Odisha and Gujarat. The low-cost cage farming technology for cobia has been well accepted by the fishermen groups and entrepreneurs in several maritime states of India. In this context, fish disease prevention and management is a very crucial aspect for successful aquaculture outcomes.

#### Diseases in cage culture of cobia

Fish cultured in floating cages become particularly susceptible to disease when various environmental parameters such as temperature, salinity, dissolved oxygen and suspended particles fluctuate suddenly or widely, or following certain cyclic climatic conditions and handling. The prevalence and spread of diseases in marine fish farming has gained more traction in recent years. The major diseases in mariculture are caused by a wide range of infectious organisms, including bacteria, viruses, fungi, protozoan and metazoan parasites and also nutritional and environmental problems including harmful algal blooms. Among the diseases in marine cultured finfish, vibriosis is a serious bacterial diseases characterized by exoptholmas, haemorrhagic gastritis, ascites, septicaemia and mortality (Rameshkumar et al 2017). After end of the nursery phase the fingerlings of cobia (each weighing 50-80g) will be stocked in the open sea cages for farming. Fish cultured in floating cages become particularly susceptible to vibriosis caused by Vibrio alginolyticus, V.parahaemolyticus and V.harveyi, whenever the juveniles are getting immunosuppressed or its defence mechanism is lowered due to any kind of stressful conditions. The development of a suitable vaccination programme plays an important role in better health management practices. As use of most of the antibiotics are banned in aquaculture, the alternative is to maintain fish health based on supplements of probiotics, immunostimulants and administration of vaccines. Mortality details of cobia in cage culture at Gulf of Mannar and Palk Bay are listed in Table1.

#### Vaccine development

The first fish vaccine was developed for enteric red mouth (Yersinia ruckeri) in salmonids during the late 1970s. The first vaccine for prevention of vibriosis in salmonids was available in 1988 by Norvax®Vibriose (Intervet, Bergen, Norway). A killed bacterial vaccine (bacterin) is currently available for V. anguillarum and V. ordalii which has been demonstrated as effective in prevention of vibriosis in juvenile Atlantic halibut as well as for salmon. The formalin inactivated vaccines were superior than heat killed preparations, especially when the bacterins were injected with adjuvants. The vaccination strategy is a better choice to control the infection caused by vibriosis. Currently vaccines against V.alginolyticus are mostly made from sonicated and heat killed bacteria (Cheng, 2009) and it is reported that cobia has developed protective immunity through vaccination with inactivated Vibrio bacterins. Lin et al., (2006) developed vaccine for cobia using a polyvalent preparation comprising inactivated V.alginolyticus, V.parahaemolyticus and P.damselae subsp. piscicida, which induced appearance of specific antibody, one week after post-injection and was detected until the end of the trial at 6<sup>th</sup> week. The fish immunized

Year	Month when mortality occurred	Mortality (in numbers)	Total length (cm) range of affected fishes	Total weight of affected fishes	Diagnosis	Type of fish affected	Location
2010	August, September	670	23.5 to 43.0	73.5 to 650 g	Vibriosis	First time cage culture of cobia fingerlings.	Mandapam
2011	June, July, August	16	81 to 135	5 to 33kg	Septicaemia due to Enterobacteriacae sp and vibriosis	Brood stocks	Mandapam
2012	May, July, August	34	94 to 110	11.1 to 18.1 kg	<i>Trichodesmium</i> bloom followed by vibriosis	Sub-adults and brood stocks	Mandapam
2013	July ,August	91	90 to 102	8 to 10 kg	Typical vibriosis	Sub-adults	Mandapam
2014	December	400	16 to 19	20 to 25g	Higher stocking density followed by vibriosis	Fingerlings.	Private sea cage- Mandapam
2015	April, July, August	183	35.5 to 43.0	41.5 to 70.5 g	Higher sea surface temperature, and Vibriosis.	Fingerlings.	Private sea cage- kattumavadi and Munaikadu
2016	March, April, May, July	1775	22 to 25.5	45 to 55g	Vibriosis, and, Viral Nervous Necrosis(VNN)	Fingerlings.	Private sea cage Thangachimadam and Munaikadu
2017	July August, September	5	102 to 117	10.0 to 13.5 kg	Nonspecific	Sub adults	Mandapam
2019	March& April	62	99 to 122	8.5 to 22 kg	Photobacterium spp. due to higher sea surface temperature	Sub-adults and brood stock in sea cage.	Mandapam
2019	September	35	110-125	14.5 to25 kg	<i>Noctiluca scintillans</i> bloom	Fingerlings and sub-adults	Mandapam

either through intramuscular or intra-peritoneal vaccine use, showed protection against challenges. Bacterial vaccination was not reported earlier in sea cage culture of marine finfishes in India. Hence, a study to the develop suitable vaccine against Vibriosis in finfish was initiated with objectives of developing a multivalent whole cell inactivated vaccine against vibriosis, standardising the dose, route and its efficiency through in-vivo experiments and evaluating the protective effect of vaccine in field conditions. The three vibrio organisms (V.alginolyticus, V.parahaemolyticus and V.harveyi) were selected for multivalent vaccine preparation based on the repeated outbreaks of the vibriosis in cobia cage culture. Formalin inactivated multivalent vibrio vaccine with aluminium hydroxide (ALGEL) as adjuvant was formulated. Only those cobia fingerlings and growout stages that had not been exposed to any microorganisms and shown to be free from specific antibodies against any systemic infections were selected for the experiments where the whole cell inactivated multivalent vibrio vaccine against vibriosis was evaluated.

Twelve cobia fingerlings each weighing about 30-32 g were alloted in three groups in triplicates (n=110), namely Group I -vaccinated, Group II- Only aluminium hydroxide adjuvant and Group III-Control Phosphate Buffered Saline (PBS). For the vaccine trial, blood was collected from the caudal vein of six cobia fingerlings at 7, 14, 21 and 42 day post vaccination (DPV). At each sampling, fish were anaesthetized with a 60  $\mu$ L/L dose of clove oil by dip for blood collection. The blood collected using a 1 mL tuberculin syringe was immediately transferred to an eppendorf tube was left undisturbed for two hours till the straw coloured serum separated out. This was



Fig.1. Intraperitoneal vaccination of cobia with a Manual vaccinator

collected by centrifugation at 3500 rpm at 4°C for 10 minutes and stored at -20°C until use.

During the challenge study, six fish from each group were challenged on the 42 DPV by intraperitoneal inoculation of 0.1 mL of *V.alginolyticus* 1x10<sup>8</sup> cfu/mL (1x10<sup>7</sup> cfu/ fish) cell suspension (Fig.1). The clinical signs, lesions and cumulative mortality were recorded daily for two weeks of post challenge and necropsy was conducted on dead fish to determine the cause of death. Re-isolation and the presence of *V.alginolyticus* in the tissues were determined by bacterial culture in the TSA and TCBS agars. The vaccine efficiency was evaluated by challenge methods in cobia fingerlings. In the challenge studies, all vaccinated fish survived without showing any clinical signs but in the adjuvant group (Group II ) 90% mortality with 10% survival and in PBS control(Group III) 100% mortality were observed at 96 h post challenge.

The relative percent survival (RPS) was calculated (Dehghani et al., 2012).

$$RPS = 1- \left\{ \frac{\% \text{ Mortality of vaccinated group}}{\% \text{ Mortality of control group}} \right\} x100$$

The post vaccination immune response was detected by ELISA and antibacterial antibody titre or the OD values were analysed following the method of Gudmundsdottir *et al.* (2009). The fish serum antibody showed an increasing trend on 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day of post vaccination (DPV) and serum antibody levels showed a decreasing trend and thereafter the immune response was maintained upto 6 weeks. The post infective response was detected by



Fig.2. Vaccinated cobia fingerlings before stocking in the cage



Fig. 3. Intra-peritoneal administration of multivalent vaccine to cobia sub-adults in cages

ELISA and the significant change (p < 0.05) of antibody response was noted in the vaccinated fish when compared to the control and adjuvant group placebo groups. (Figs. 3, 4 & 5)

After the standardization of multivalent vibriosis vaccine in *in-vitro* and experimental study, the field application trial was carried out in the sea cage farmed cobia fishes. Cobia sub adults (90 numbers) and brood stock (35 numbers) were vaccinated with multivalent (*Vibrio alginolyticus, V. parahaemolyticus* and *V. harveyi*) Algel adjuvant vaccine during July and September 2017. One booster dose was also given after 35 days of the first dose.

Among the six cages, one cage grow out fish (30 numbers) were kept as control without vaccination. ELISA was performed to identify the serum optical density (OD) levels in the control and the vaccinated groups. The cobia serum antibodies showed rising trends from 7<sup>th</sup> Day of Post Vaccination (DPV) and extended upto 21 DPV. Then the serum antibody OD levels showed decreasing trend after 21 DPV and serum antibodies were detected upto 6 weeks. The OD values of antibody to multivalent vaccine differed significantly (p<0.05) in the laboratory and in the field trial. There was a significant (p<0.05) increase in the OD values of antibodies from 7<sup>th</sup> to 21<sup>st</sup> day and dropped significantly (p<0.05) at 28 DPV. Hence, a



Fig. 4. Hyper-immune serum collected after 35 DPV

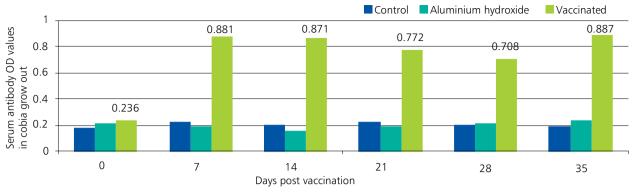


Fig.5. Hyper-immune serum analysis by ELISA

booster on 28<sup>th</sup> DPV was administered. On the 35<sup>th</sup> day OD value was higher than 21<sup>st</sup> and 28<sup>th</sup> day, indicating that serum antibody OD levels were increasing after the booster dose of the vaccine and the immune response was extended upto further 6 weeks. (Figs.4 & 5)

The first vaccination was done during April 2017 followed by a booster dose in July 2017 for the growout and broodstock cobia fishes cultured in Gulf of Mannar. No epizootics were observed during the culture period of one year. Again the next vaccination was initiated in April 2018 followed by the booster dose during July 2018. Even though the seasonal blooming (*Trichodesmium* sp) occurred during August and September, the vaccinated fish didn't show any stress condition or succumb to any diseases. But >60% mortality was observed in the control, unvaccinated fishes in the control cage. The same vaccination schedule was again followed during April 2019 with booster dose in July 2019 and vibriosis incidence was averted.

#### Standard guide lines

The study was performed based on the European medicines agency's standard guidelines 2013. To assess the acute safety characteristics of the vaccine, the fish should be monitored daily for mortality/ morbidity over a minimum of a 14<sup>th</sup> day period. For parenteral vaccine, necropsy examination should include investigation of the occurrence of effects such as pigmentation (Eg. Melanization ) and adhesions measured using the 'Speilberg score' (Midtlyng *et al.* 1996). The mortality is an evaluation parameter in vaccine challenge study. In our experimental finding the pathomorphological lesions were within the score of 0 and 1 grade. There was no visual appearance of lesions in abdominal cavity and no any minor opacity of peritoneum after evisceration.

#### Conclusion

The main objective of developing a multivalent vaccine against vibriosis is to prevent the seasonal epizootics in sea cage farmed cobia. It is concluded that in cobia fingerlings and growout fishes, the immune response can be improved against vibriosis by the timely vaccination. This might be due to the well developed detectable serological antibodies against vibriosis (Evaluated by challenge studies in fingerlings and ELISA) and establishment of acquired immunity by the vaccination method during sea cage farming activities. The acquired immunity might have developed after the vaccination, reduced the chances of life threatening infections. Thus, the regular epizootics observed in cage culture cobia, in every year during the month of July to September (pre-monsoon season) were prevented by vaccination and proper sea cage farming management. The vibriosis incidence has been successfully controlled by proper vaccination schedule. In sea cage farming of cobia, the first vaccination at the hatchery and subsequent booster injection at the cage will give the elevated antibody titres and immune response for more than 3 months. So, the fisherman or the cage farmers would not get any losses during the entire culture period, due to the seasonal epizootics of vibriosis.

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Taylor *et al.*, 1998. *Aquaculture*, 162: 219-230. (Reference with more than two authors)

Friedman and Bell. 1996. *J. shellfish Res.*, 15: 535-541. (Reference with two authors)

Pauly, 1980. FAO Fish. Tech. Pap., 234.

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