

Methodology

Study Area

Gujarat state nestled in the westernmost part of India, emerges as a coastal treasure, flaunting an expansive shoreline that stretches over a vast length of approximately 1,600 kilometers. This coastal zone not only holds the distinction of being the longest among all the coastal states in the country but also accounts for an impressive 21% of India's entire coastline.

Within the enchanting realm of Gujarat, the South Saurashtra coastline takes centre stage in the realm of the research. Spanning a generous 850 kilometers, this coastal stretch is oriented towards the west, presenting a mesmerizing tapestry of land and sea that beckons exploration and investigation for this research. After careful consideration three distinctive locations viz. Mangrol ($21^{\circ} 4.2' N, 70^{\circ} 4.2' E$), Adri ($20^{\circ}57'35.1'' N, 70^{\circ}16'43.0'' E$) and Veraval ($20^{\circ}54'37'' N, 70^{\circ}21'04'' E$) were chosen, each offering unique characteristics that make them ideal for this study.

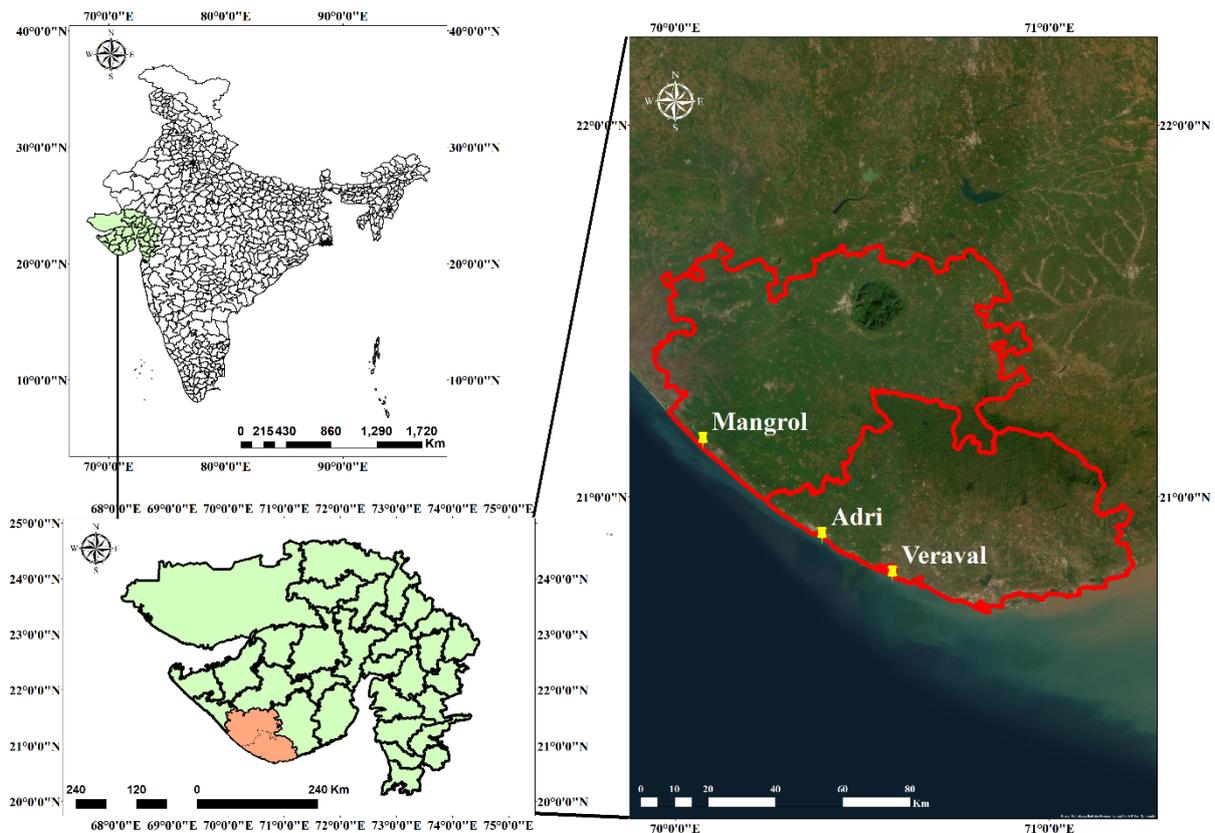


Fig. Study area

Mangrol:

The Mangrol coast lies at latitude 21°07' N and longitude 70°07' E, situated on the southwestern coast of Gujarat within the Junagadh district. This coastal region boasts an extensive rocky intertidal zone, creating a conducive sheltered ecosystem for various mollusc species and other fauna. The Mangrol coastline can be divided into two distinct sections flanking Juni-Jetty, with a combined length of approximately 7.5 to 8 kilometres. The left side of Juni-Jetty stretches for about 4 kilometres and exhibits a less anthropogenically influenced intertidal zone. Conversely, the right side, extending approximately 3.5 to 4 kilometres from the Juni-Jetty, is more affected by human activities (Gohel 2016). This area is primarily used by fishing communities for activity such as fish bait collection during low tide. Additionally, it serves as a disposal site for fishing waste, damaged nets, discarded plastics, and boat parts, adding to its multifaceted characteristics and utilities. It is due to these unique attributes that this study site was chosen. Regarding site characteristics, the Mangrol study site is characterized by a predominantly rocky intertidal coastline, occasionally interspersed with sand and notably devoid of mud or silt. A clear demarcation exists between the sandy areas and the rocky substratum, marking an important coastal feature. Furthermore, small water bodies such as pools and puddles in the area have their bottoms filled with sand. The rocks in this region typically consist of calcareous material, with the consolidated ancient equivalent known as milliolite rocks.

Adri:

Adri, situated along the southern part of the Saurashtra Coast, boasts a shoreline spanning approximately 1.5 to 3 kilometers in length. This coastal stretch predominantly features a rocky shore composition, occasionally punctuated by sandy patches, yet notably lacks steep inclines or abrupt changes in terrain. Within the selected areas, the intertidal zone extends over a range of 60 to 70 meters. Anthropogenic activities were observed to some degree across all the designated sampling sites, with a noticeable increase in such activities attributed to the presence of a temple in Adri. It is worth noting that the coast's unique combination of rocky and sandy elements, coupled with its relatively straight and gentle terrain, renders it an ideal habitat for conducting intertidal ecological studies, free from steep gradients or abrupt changes in topography.

Veraval:

Veraval, situated on the southwest coast of Saurashtra, lies approximately 6 kilometers from the revered Somnath Mahadev Temple, a site of great importance in Hindu mythology as it is considered the first of the 12 Jyotirlingas. Geographically, Veraval is positioned at a latitude of 20° 54' 37" N and a longitude of 70° 21' 04" E, with the city's elevation averaging at sea level. Notably, Veraval is renowned for its flourishing fishing industry, being one of the largest fish landing centers in India. The "Kharwas" fishing community holds significant influence in Veraval's fisheries governance. Veraval is renowned for its boat construction and seafood industry, extending to global markets like the USA, Europe, and Asia. Our study site, chosen near Veraval Fisheries College and Chowpati lighthouse, is strategic for sampling. During low tide, an area spanning approximately 30 to 50 meters of the intertidal zone becomes exposed. The intertidal region is characterized by a composition of roughly tidepools, crevices, and flat terrain. Notably, the majority of shallow tidepools are concentrated in the upper and middle intertidal zones, while the lower intertidal region features larger tidepools and a steep, vertical slope leading towards the subtidal zone. Beyond the upper intertidal region lies the spray zone, which primarily consists of sandy shores. The rocky shoreline exhibits discontinuities, often interrupted by large boulders or deep channels.

Time Span:

The research was conducted over a broad duration, spanning from February 2021 to March 2023, to comprehensively investigate the molluscan diversity and population ecology of the molluscs in the selected study sites. The research methodology commenced with a preliminary pilot survey conducted from February 2021 to April 2021, primarily focused on site selection and the identification of suitable rocky intertidal zones for the study. Upon the completion of the initial survey, the specific sampling sites were delineated, paving the way for the subsequent phases of the research. A systematic and consistent monthly visitation schedule to the chosen study sites commenced, which spanned approximately one year, starting from April 2021. During this phase, the research primarily focused on studying the diversity of molluscan species within the intertidal zone. The data collection of population ecology began in November 2021 and

extended through October 2022, marking an essential aspect of the research. Concurrently, comprehensive length and weight data of selected species were also gathered during this period. Importantly, these visits to the selected study sites were diligently maintained monthly, culminating in the accumulation of substantial datasets. Furthermore, to ensure the accuracy and reliability of the research findings, an additional phase of visits, until March 2023, was carried out at monthly intervals. The objective of this extended phase was to compile a comprehensive database encompassing the presence and characteristics of molluscan species. The data collected during this period were meticulously cross-checked against the previous year's data, facilitating a thorough verification of the findings of study and their scientific validity. Throughout the research process, a standardized approach was adopted for surveying all selected study sites, with a particular emphasis on conducting surveys during the lowest tides to maximize data collection opportunities. The identification and scientific classification of observed intertidal molluscan species were conducted systematically and accurately. Furthermore, the research findings were rigorously validated through consultation with various scientific resources, including bulletin boards and online forums, as well as by referencing relevant research publications and thesis available on the internet.

Habitat Characterization

To achieve the primary research objective and aim of investigating molluscan diversity on the South Saurashtra coast, a detailed assessment of habitat characterization was performed. This involved the identification of various habitats from selected sampling stations along the South Saurashtra shoreline. The study, focusing specifically on molluscan species, was systematically conducted across three distinct seasons: winter, summer, and monsoon, to ensure a comprehensive understanding of seasonal variations in molluscan distribution and habitat preferences. This seasonal approach was essential to capture the full spectrum of environmental conditions influencing molluscan ecology in this region.

Sampling Method for Molluscan diversity

To investigate the diversity of molluscan species, comprehensive field surveys were conducted from February 2021 to October 2022, employing a systematic hand-collection

approach. Throughout the fieldwork, distinctive morphological characteristics of the molluscs were meticulously noted, and photographs capturing their unique coloration and shell patterns were taken for documentation purposes. Following collection, the specimens were carefully preserved in 70% ethanol and transported to the Division of Marine and Freshwater Biology Laboratory at the Department of Zoology, The Maharaja Sayajirao University of Baroda (MSU). Upon arrival at the laboratory, the specimens were sorted based on distinguishing features such as shell shape, texture, and aperture characteristics. Identification to the species level was carried out by comparing their morphological features against established illustrative keys, relevant research papers, and authoritative monographs. This step was crucial in ensuring accurate classification and understanding the diversity within the molluscan community. To supplement and validate the species identification, all specimens were cross-referenced with images and identification characteristics available on renowned marine biology databases and websites. The most recent taxonomic classification and nomenclature of the molluscs were adopted from the World Register of Marine Species (WoRMS) website (www.marinespecies.org), ensuring the study aligned with contemporary scientific standards. For the collected molluscan specimens, various morphometric measurements were conducted, including shell length and shell weight.

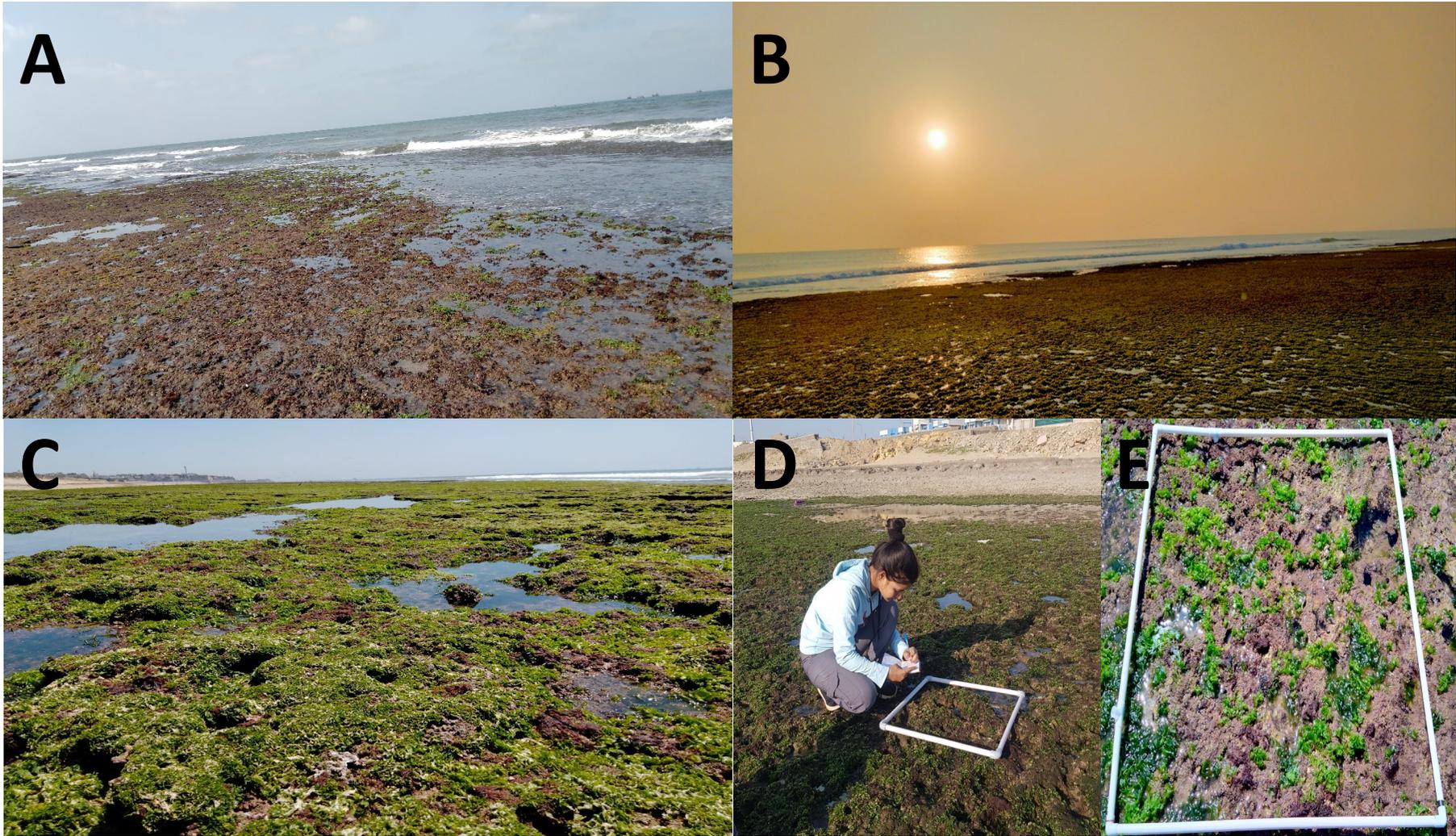


Figure: Habitat of study area (A) Mangrol (B) Adri (C) Veraval (D) & (E) Quadrata sampling method

Species Distribution and Ecological Status

The distribution patterns of molluscan species along the South Saurashtra coast were examined by focusing on various substrata and zonations, including the upper, middle, and lower intertidal zones. This analysis was conducted to understand the spatial and temporal variations in molluscan populations across these distinct ecological niches. By estimating a range of population indices, the study aimed to discern how different environmental factors in these zones influence molluscan distribution. Additionally, the investigation into seasonal variations in population dynamics within these specific zones was conducted to assess the ecological status and adaptability of molluscan species in response to changing environmental conditions along the South Saurashtra coastline.

Zonation

The intertidal zone, also known as the littoral zone, is a unique marine environment characterized by its exposure to air at low tide and submersion at high tide (Leeuwis and Gamperl 2022). This zone stretches between the lower and higher tidal marks and is remarkably distinct for its harsh conditions, influenced by both terrestrial and marine factors. Within this zone, there exists a fascinating phenomenon called intertidal zonation, which refers to the vertical stratification of organisms (Raffaelli and Hawkins 1996). This stratification is particularly evident in the molluscan communities, which adapt to the varying conditions of the intertidal subzones: the spray zone, high intertidal zone, middle intertidal zone, and lower intertidal zone (Knox 2000). Mollusca, a diverse phylum in the marine ecosystem, exhibits remarkable adaptations to the intertidal environment (Harkantra, Rodrigues et al. 1982);(Mettam 1994). The ecological pressures such as wave action, tidal cycles, and temperature fluctuations significantly influence the distribution and behaviour of molluscan species across the different intertidal subzones. For instance, in the spray zone or the supra-tidal zone, molluscs that can withstand desiccation and high salinity variations are predominant (Struhsaker 1968). These species often have robust shells and specialized physiological mechanisms to conserve water. Moving to the high intertidal zone, or the upper littoral zone, molluscs here are adapted to longer periods of air exposure and less frequent submersion. They often possess mechanisms for moisture retention and can withstand temperature extremes. The middle intertidal zone, or the mid-littoral zone, hosts a greater diversity of molluscs, benefiting from more regular submersion and reduced exposure to the harsh terrestrial

environment (Boaden and Seed 1985). Molluscs in this zone display adaptations such as strong attachment mechanisms to withstand wave action. The lower intertidal zone, or the lower littoral zone, is the most marine-influenced zone with molluscs that are adapted to almost continuous submersion. Here, the species are less adapted to desiccation but are often more specialized for dealing with predation and competition in a more consistently marine environment.

Spray Zone

The Spray Zone, situated beyond the spring high-tide line, is an ecologically distinct area that is typically submerged only during extreme weather events, such as severe storms, or exceptionally high tides. This zone, characterized by its relative dryness and sparse population, presents an environment where fluctuations in moisture, temperature, and salinity are more pronounced compared to other intertidal zones (Vaghela 2010). Given these conditions, the Spray Zone is often considered to be more terrestrial than marine in nature. Despite its challenging environment, the Spray Zone does support a unique assemblage of flora and fauna, including certain molluscan species. The molluscs found in this zone have evolved specific adaptations to thrive in such a variable habitat. They are exposed predominantly to the atmospheric conditions and only occasionally to marine influences, primarily in the form of wave splashes and wind-driven spray (Mustoe 2010). The molluscs inhabiting the Spray Zone exhibit unique physiological and behavioural adaptations. These adaptations might include highly efficient moisture retention mechanisms, the ability to tolerate a wide range of salinity, and the capacity to endure significant temperature fluctuations. The shells of these molluscs are often robust, providing physical protection against desiccation and temperature extremes (Marin, Luquet et al. 2007). Furthermore, these organisms may possess specialized reproductive strategies to ensure the survival of their species in such a sporadically inundated environment (Sabelli and Taviani 2014).

Upper Intertidal zone

The upper intertidal zone, a unique marine-terrestrial interface, is primarily submerged during high tides and exposed to the air for extended periods during low tides. This zone experiences the highest tides but predominantly functions as a terrestrial habitat due to its prolonged exposure to air. The environmental conditions here are characterized by

limited water availability, which poses a significant challenge for sustaining diverse marine life (Leeuwis and Gamperl 2022). Despite these challenges, a variety of organisms have adapted to survive in this fluctuating environment. Among them, molluscs such as chitons and limpets, along with barnacles, crabs, and green algae, are the predominant species. These organisms demonstrate remarkable adaptations to cope with the varying levels of submersion and exposure. Molluscs in the upper intertidal zone possess specialized features to endure prolonged periods of desiccation (Veras, Martins et al. 2013). For instance, chitons and limpets have evolved strong attachment mechanisms, allowing them to cling tightly to rocks, thereby reducing water loss during low tides. Their shells are designed to minimize water evaporation, and their metabolic activities adjust according to the tidal rhythms to conserve energy and resources. Rock pools, a common feature in this zone, provide temporary refuges where water is retained during low tides (Smith 2013). These pools act as microhabitats, supporting a more diverse range of molluscan life by offering a more stable environment compared to the surrounding zone (Gonçalves, Vinagre et al. 2023). The upper intertidal zone, with its distinct environmental conditions, supports a more abundant and diverse life compared to the spray zone. The molluscan species here are a testament to the adaptive capabilities of marine organisms, showcasing their resilience and ability to thrive in a habitat that oscillates between marine and terrestrial conditions.

Middle Intertidal Zone

The middle intertidal zone, situated between the high tide and low tide marks, experiences a unique environmental condition where it is submerged underwater and exposed to the air for approximately equal durations during each tidal cycle. This zone encounters more vigorous wave action compared to the upper intertidal (high tide) and spray zones. As a result, the middle intertidal zone harbours a diverse range of organisms, distinguishing it significantly from the relatively less diverse splash and high tide zones. In terms of molluscan life, the middle intertidal zone is particularly diverse and abundant. This zone supports a variety of molluscs, such as *Onchidium*, a notable gastropod, along with other marine organisms including anemones, barnacles, crabs, various algae, and sponges (Satyam and Thiruchitrambalam 2018). The molluscs in this zone have evolved specialized adaptations to cope with the frequent changes in their environment due to tidal actions (Newell 1976). These adaptations include physiological mechanisms to

tolerate fluctuations in salinity and temperature, as well as structural features that aid in withstanding strong wave impacts. The richness in marine vegetation and fauna in the mid intertidal zone leads to a highly differentiated ecosystem, contrasting markedly with the ecosystems of the high tide and spray zones (Raffaelli and Hawkins 1996). The presence of diverse molluscan species in this zone reflects their adaptive capacities to thrive under the dynamic conditions of regular submersion and air exposure.

Lower Intertidal Zone

The lower intertidal zone, primarily submerged and only exposed during low tides or exceptionally low tides for extended periods, is characterized by a high density of marine life. This zone is distinct from the upper intertidal regions due to its abundant marine vegetation, particularly seaweeds, which thrive here more than in the other subzones. For molluscs, the lower intertidal zone provides a unique habitat. The richness of this zone in species is attributed to the fact that organisms here are submerged most of the time, fostering more interactions among different species compared to other intertidal zones. Molluscs in this region, such as various snails, are not adapted to prolonged periods of water scarcity or high temperatures, unlike their counterparts in the higher zones (Newell 1976). The flora and fauna, including seaweeds, crabs, algae, sea stars, sea urchins, shrimp, snails, and sponges, benefit from the consistent water availability. This constant marine environment allows for larger growth and development, particularly in marine plants. The ample water supply ensures that enough light penetrates for photosynthesis, while maintaining near-normal salinity levels, both of which are crucial for the thriving of marine vegetation (Park, Kim et al. 2016).



Figure. Displaying distinct intertidal zones 1) Upper intertidal Zone 2) Middle intertidal Zone 3) Lower intertidal Zone

The selection of Dominant species

The selection of dominant species for the study was meticulously executed based on their prominent presence and abundance in the chosen research area. These organisms, consistently noticeable and prevalent in the intertidal zone throughout the year, were identified as non-migratory (inter-coastal), ensuring the feasibility of extended research on a consistent subject. It was also observed that these species exhibited dominance over other species present across all three zones of the intertidal area at the selected sampling site. During the survey, certain mollusc species were observed to be particularly abundant in the selected rocky intertidal zone. Consequently, for the purposes of studying population ecology, these mollusc species were chosen. This approach not only facilitated a focused examination of the intertidal ecosystem but also allowed for a comprehensive understanding of the ecological dynamics and species interactions within this habitat.

Table 1: - Taxonomy of Selected Dominant species

Phylum	Class	Order	Family	Genus	Species
Mollusca	Gastropoda	Caenogastropoda	Cerithiidae	Cerithium	<i>Cerithium caeruleum</i>
		Turbinidae	Turbinidae	Lunella	<i>Lunella coronatus</i>
		Onchidiacea	Onchidiidae	Peronia	<i>Peronia verruculata</i>
		Trochida	Trochidae	Trochus	<i>Trochus radiatus</i>

Quadrat Sampling

Quadrat sampling was systematically conducted in a zig-zag pattern, strategically covering the maximum exposed intertidal zone to comprehensively study the population ecology of selected molluscan species and the size of the quadrates laid was 1 m² (Appukuttan, Chellam et al. 1989). The collection area was precisely determined at fixed time intervals, employing a hand-picking method. Quantifying the abundance of molluscan fauna was achieved by converting the collected data into individuals per square meter through appropriate calculations as outlined by Ksyunin (2000). The intertidal survey was meticulously carried out twice a month throughout the entire study period, ensuring a comprehensive understanding of temporal variations. Any gaps in observations were addressed through random visits over the following two years. Visual observation and photo-interpretation were integral components of the methodology, ensuring a thorough examination of each quadrat. This approach facilitated a robust assessment of molluscan distribution and abundance in relation to the varying environmental conditions along the intertidal gradient.

Zone wise distribution of species

Sampling was performed during low tide, ensuring access to all three intertidal zones: upper, middle, and lower. Field observations were conducted during low tide to

ensure access to all three intertidal zones. Mollusc species observed in different zones were noted directly in the field.

Species Distribution Model

For the purpose of this SDM study, specific sites along the Gujarat coast have been selected based on their prominence in Molluscan distribution (Vadher, Kardani et al. 2023). These sites fall within the coordinates ranging from approximately 20.6°N to 23.7°N latitude and 68.9°E to 72.6°E longitude. These coordinates cover critical habitats from the Gulf of Kachchh, known for its rich coral reefs and seagrass meadows, to the estuarine regions of the Gulf of Khambhat and the expansive coastline of Saurashtra, which provides a home to numerous gastropod and bivalve species (Sivakumar 2019). It will also offer insights into potential areas for expansion and intensification of Molluscan farming, providing economic benefits while ensuring ecological sustainability.

a) Background on the importance of species distribution mapping

Regular field visits were conducted to evaluate the diversity and distribution of Molluscan species along a designated coastal area. During these investigations, a total of 60 Molluscan species were identified, with a collective count of 3,261 individual organisms being recorded. From this extensive data collection, four dominant species were distinguished based on their dominance and abundance, highlighting the necessity for focused farming strategies namely *Cerithium caeruleum*, *Lunella coronatus*, *Peronia verruculata* and *Trochus radiatus*. The primary aim of this research paper is to highlight the pivotal role played by species distribution mapping in the efficient cultivation of molluscs. The elucidation of specific habitat preferences and distribution patterns of various Molluscan species is shown to make a significant contribution towards the optimization of Molluscan aquaculture practices. This optimization is facilitated by arranging farming locations with the Molluscs natural ecological needs, thus improving their chances of survival, growth rates, and overall productivity of the farms. Furthermore, the paper explores the wider ecological ramifications of species distribution data, especially in terms of understanding the effects of environmental changes on Molluscan communities. The meticulous monitoring of changes in species distribution in reaction to environmental factors such as water temperature, salinity, and chlorophyll a concentration is discussed. The species distribution models (SDMs) are

identified as crucial in protecting Molluscan aquaculture operations from environmental challenges, thereby enhancing their sustainability and resilience amidst shifting ecological conditions.

b) Species sampling and identification techniques.

Quadrat sampling stands as an advantageous technique employed in the study of Molluscan species along the Gujarat coast, offering a systematic and quantifiable method for species sampling and identification (Wells, Chalermwat et al. 2008). This method involves laying out square plots of a set size, known as quadrats, at regular intervals across the study area to ensure a representative sample of the Molluscan population is assessed. The use of quadrats is particularly beneficial in delineating the distribution of stationary or slow-moving organisms, such as many molluscs (HAAG, DISTEFANO et al. 2012). It facilitates the precise recording of species presence, abundance, and spatial distribution, enabling researchers to generate accurate data that is crucial for effective species distribution modelling (SDM). Additionally, quadrat sampling allows for the comparison of data across different habitats and time periods, making it a robust tool for monitoring environmental changes and their impact on Molluscan communities. By adopting this technique, the research on the Gujarat coast provides reliable and repeatable results that are integral to understanding and managing the region's diverse Molluscan populations.

c) Occurrence data collection methods.

For the collection of occurrence data of Molluscan species along the Gujarat coast, a systematic approach was employed to ensure comprehensive and accurate representation of the distribution of species. Field surveys were meticulously planned to coincide with seasonal cycles and tidal patterns, which are known to influence Molluscan activity and visibility (Tran, Nadau et al. 2011). The Research conducted transect walks and utilized quadrat sampling methods, where predetermined square plots of specific dimensions were laid out at regular intervals along various habitats of the coastline. This approach allowed for the standardized collection of data on the presence and abundance of Molluscan species.

During these surveys, detailed records of each Molluscan specimen encountered were kept, noting the species, size, and distinctive features. garmin gps etrex 10 device was

used to record the precise locations of each sighting, providing georeferenced data points that are crucial for the creation of accurate distribution maps. In addition to field observations, local fishermen and other stakeholders were interviewed to gather ancillary data on less accessible or deeper coastal areas. Specimens collected were taken to laboratories, Division of Marine and freshwater biology, Department of Zoology, The Maharaja Sayajirao University of Baroda and Zoology Lab, Bhakta Kavi Narsinh Mehta University for further identification and validation, especially when dealing with cryptic or juvenile forms, to augment the field identification process. The accumulated data from these various methods form a robust dataset for analysing the distribution patterns and habitat preferences of molluscs on the Gujarat coast, providing a solid foundation for ecological studies and the development of conservation strategies.

d) Quadrature studies for key species selection and data collection

Quadrature studies for key species selection and data collection are central to understanding the distribution of Molluscan species along the coast. This methodology proved to be instrumental in identifying the dominant species within the ecosystem. Post-analysis, it was discovered that four species notably stood out due to their prevalence. This prominence was determined based on the ratio of individuals of the most abundant species relative to the total population of molluscs in the sampled ecosystem. Dominant species are typically those that have a significant impact on the community structure and the distribution of other organisms within the same habitat. The degree of dominance among different communities or samples, particularly when the number of species and total abundances vary in the study area is calculated by Whittaker's index as shown below in Eq1.

The formula for calculating Whittaker's Index is:

$$I\delta = \log_{10}\left(\frac{N}{n}\right) \dots\dots\dots (1)$$

Where:

N = Total number of individuals in the sample

n = Number of individuals of the most abundant species

In the scope of this study, Whittaker's Index (I_δ) is a measure of dominance that quantifies the degree to which the most abundant species dominates the community relative to the number of species present (CASTRO and JAKSIC 2008). In the study area, which spans the upper, middle, and lower intertidal zones, 4 out of 60 species were identified as dominant. The species that stood out due to their prevalence are *Cerithium caeruleum* with a mean value of 0.066, *Lunella coronatus* at 0.056, *Peronia verruculata* with 0.074, and *Trochus radiatus*, which had the highest mean value of 0.083. Their high numbers not only illustrate their successful adaptation to local environmental conditions but also their potential impact on the ecological dynamics of the region. Understanding the abundance and spatial distribution of these species provides invaluable insights into the health of the ecosystem and aids in the socio-economic development. The prevalent presence and potential ecological resilience of these dominant species might also suggest their viability for aquaculture projects in the area.

e) Acquisition and processing of environmental data from Bio-Oracle

The acquisition and processing of environmental data from Bio-Oracle underpin the ecological assessments and predictive modelling for marine species distribution, including molluscs (Bolam, Cooper et al. 2023). Bio-Oracle is a comprehensive marine data repository that offers a wide array of global environmental layers which are crucial for Species Distribution Models (SDM). These layers typically include various oceanographic and biotic variables, such as sea surface temperature, salinity, and chlorophyll -a concentration levels, which are often provided at high spatial resolution as depicted in table 2.

Table 2: Selected environmental predictors suitable for benthic species distribution modelling along with their biological importance.

Predictor	Unit	Biological Importance
Mean Surface Salinity	pss	Salinity is used to define different water masses and depth zones and is considered as a primary driver for the distribution of benthic invertebrates(Russell, Connell et al. 2012)
chlorophyll -a concentration levels	mg/m3	Primary productivity proxies indicate food availability for suspension feeding mollusks (Rodil, Compton et al. 2014)
Mean sea surface temperature	c	Temperature is a limiting factor for marine species distribution that controls metabolic rates and affects physiological functions in all growth stages(Velaoras, Kassis et al. 2013)

To utilize the Bio-Oracle for SDMs, first acquired relevant environmental data layers that align with the scope and scale of their study. This usually involves selecting variables known or hypothesized to influence the distribution of the target Molluscan species. Once these layers are downloaded, the data undergo processing which might include clipping to the study area's spatial extent, ensuring compatibility with other data sets, and statistical analyses to discern patterns and correlations. Processing also involves cleaning the data to remove any anomalies or errors and standardizing the datasets to a common format and spatial resolution to ensure consistency across the variables. The quality and resolution of these data layers are paramount, as they can significantly impact the predictive accuracy of the SDMs. With properly processed environmental data from Bio-Oracle, it can then correlate the presence or absence of Molluscan species with

environmental conditions, leading to robust predictive models that can inform sustainable socio economy spots for mollusc populations along coastlines of the study area.

f) Methodologies employed for predictive mapping.

In ecological studies, predictive mapping is essential for understanding species distribution patterns, and four distinctive modelling approaches are commonly utilized to achieve this, each offering unique advantages and mechanisms suitable for various types of data. The Maximum Entropy Model, known as Maxent, is founded on the maximum entropy principle (Wiltshire and Tanner 2020). It excels in predicting species distributions using incomplete data by estimating the widest possible probability distribution of species occurrences within the given constraints. Maxent proves especially advantageous when dealing with presence-only data, as it does not rely on absence information, making it a robust tool for modelling the distribution of rare or elusive species as illustrated in Fig. 2.(Flowchart in objective 3 chapter)

The BIOMODelling framework, or BIOMOD, is a sophisticated R-based system designed for ensemble forecasting that incorporates a multitude of species distribution models(Li, Donizelli et al. 2010). It works with both presence-absence and presence-only data, enabling users to cross-validate and compare outcomes from various modelling methods like generalized linear models, generalized additive models, and classification trees(Thuiller, Lafourcade et al. 2009). BIOMOD's ensemble method amalgamates multiple predictions, yielding more precise and confident projections that are crucial for conservation efforts and understanding the potential impacts of climate change on species distributions(Thuiller 2003). Bayesian models utilize Bayes' theorem to refine the probability estimates for hypotheses based on new information, allowing them to incorporate prior knowledge into species distribution modelling. These models are particularly valuable when historical data or expert insights are available, enhancing predictive accuracy by integrating these with current observations. Their ability to manage complex data and quantify prediction uncertainties makes Bayesian models increasingly popular in ecological and geographical research(Dormann, Calabrese et al. 2018). The Random Forest model is a robust non-parametric method that generates numerous decision trees and uses their collective outcomes for classification or regression tasks(Ho 1995). In species distribution modelling, Random Forest is adapt at

processing large sets of predictor variables and capturing intricate interactions within the data. Its high precision and provision of variable importance metrics make it an essential model for pinpointing the crucial environmental factors influencing species distributions.

The research objective is to identify the most effective algorithm Species Distribution Model (SDM) for optimizing Molluscan farming. The hypothesis posits that among the various modelling approaches, an ensemble model that combines the predictive capabilities of Maxent, BIOMOD, Bayesian models, and the Random Forest model will yield the highest accuracy and reliability in forecasting suitable habitats for Molluscan aquaculture. This ensemble approach is anticipated to leverage the strengths of each individual model, such as Maxent's efficiency with presence-only data, BIOMOD's ensemble forecasting power, Bayesian models' incorporation of prior knowledge, and the Random Forest model's handling of complex data. The synergistic integration of these models is expected to provide a nuanced, multi-faceted view of habitat suitability that can be directly applied to improve the sustainability and yield of Molluscan farming practices.

g) Model training, testing, and evaluation methods

Model training, testing, and evaluation are critical phases in the development of Species Distribution Models (SDMs), ensuring that the models are both accurate and reliable for predicting the distribution of species such as molluscs along the Gujarat coast. During the training phase, the model is built using a portion of the collected occurrence and environmental data. This process involves adjusting the model parameters to best fit the known distribution of the species based on the selected environmental variables. Techniques such as cross-validation, where the dataset is partitioned into complementary subsets, are commonly used to train the model while avoiding overfitting(Kuhn, Johnson et al. 2013).

The testing phase involves applying the trained model to a separate set of data not used during the training phase. This step is crucial for assessing the model's predictive performance on new, unseen data, providing an indication of its generalizability and reliability in real-world applications. Various metrics, such as the Area Under the Receiver Operating Characteristic Curve (AUC) for binary classification tasks, are used to quantify the model's accuracy, sensitivity, and specificity in predicting species presence or absence(Shabani, Kumar et al. 2018). Evaluation methods extend beyond statistical

metrics and include comparing model predictions against independent occurrence records or expert knowledge to gauge the model's ecological plausibility. Model evaluation may also involve assessing the spatial patterns of predicted suitable habitats against known biological and ecological principles, ensuring that the model's outputs align with established understanding of the species' habitat requirements and behaviours. Through iterative refinement, incorporating feedback from testing and evaluation, the model is honed to provide reliable and ecologically meaningful predictions of Molluscan distribution along the Gujarat coast as given in in Fig. 2.(Flowchart in objective 3 chapter.

h) Validation procedures using ground data.

Validation of Species Distribution Models (SDMs) using ground data is an integral part of ensuring the accuracy and reliability of the model predictions. In this methodology, the model's predicted distributions of Molluscan species along the Gujarat coast are cross-referenced with independently collected ground-truth data. This ground data is obtained through field surveys and observations conducted after the model has been developed, specifically targeting areas where the model predicts high suitability for the species as well as areas of low predicted suitability to test the model's full range of predictions.

The validation process involves systematically recording the presence or absence of the target Molluscan species within these areas, using standardized sampling techniques such as quadrat sampling or transect walks, consistent with the initial data collection methods. These observations are then compared to the model's predictions to assess the congruence between predicted and observed species occurrences.

Statistical measures are employed to quantify the model's performance, including metrics such as accuracy, precision, recall, and the kappa statistic, which evaluates the agreement between observed occurrences and model predictions beyond chance. Additionally, confusion matrices may be used to provide a detailed breakdown of true positives (correctly predicted presences), false positives (incorrectly predicted presences), true negatives (correctly predicted absences), and false negatives (incorrectly predicted absences). This validation approach not only tests the model's predictive power but also highlights potential areas for refinement. Discrepancies between predicted and observed data can indicate the need for adjustments in model parameters, the inclusion of

additional environmental variables, or further investigation into the ecological dynamics of the study area. Through rigorous validation using ground data, the reliability of SDMs in predicting the distribution of Molluscan species along the Gujarat coast can be significantly enhanced, contributing to more informed conservation and management decisions as shown in Fig. 2.(Flowchart in objective 3 chapter)

Projection of models onto the Gujarat coast.

Projecting Species Distribution Models (SDMs) onto the Gujarat coast for Molluscan species involves translating the model's predictions to generate detailed spatial maps that highlight potential habitats and distribution patterns across the region. This process entails overlaying the SDM outputs onto geographical maps of the Gujarat coast, utilizing GIS (Geographic Information System) software to visualize the correlation between environmental variables and the likelihood of Molluscan presence. These projections consider the unique ecological characteristics of the Gujarat coastline, including its varied substrates, tidal regimes, and salinity gradients, which are critical determinants of Molluscan habitat suitability.

The resultant maps provide a comprehensive view of areas where environmental conditions align with the optimal habitat requirements of the target Molluscan species, identifying zones of high, moderate, and low suitability. This spatial representation allows for a nuanced understanding of the potential distribution areas, factoring in both the current state of the coast and projected changes due to factors like climate change or human activities. Moreover, these projections are instrumental in guiding conservation efforts, informing sustainable aquaculture practices, and identifying priority areas for further research and monitoring.

By integrating the SDMs with the geographical context of the Gujarat coast, researchers and policymakers can discern patterns and trends that may not be apparent from raw data alone. This approach enables the identification of habitat fragmentation, potential corridors for species migration, and areas vulnerable to environmental stressors, offering valuable insights for the management and preservation of Molluscan biodiversity in the region as shown in Fig. 2.(Flowchart in objective 3 chapter).

Gonadal Sampling

The Gonadal sampling was done of four selected dominant species. Samples were collected using appropriate methods, and gender separation was accomplished through external characteristics or, if necessary, through dissection. Gonads were carefully extracted using a sterile dissection kit, minimizing damage during the process. The extracted gonads were immediately preserved in a 10% formalin solution for at least 24 hours (Howard 2004). After fixation, the samples underwent a dehydration process using a series of ethanol solutions, followed by clearing with xylene (Peters, Price et al. 2005). The dehydrated and cleared gonads were then embedded in paraffin wax to facilitate sectioning with a microtome. Thin sections (5-10 micrometers) were mounted on glass slides and stained with Hematoxylin and Eosin for enhanced contrast. Microscopic examination allowed for the identification and documentation of reproductive structures. Histological data were analyzed to draw conclusions regarding different maturation stages (Abadia-Chanona, Avila-Poveda et al. 2016).

Data Analysis

i) Biodiversity indices

➤ Shannon weiner Index

The 'Shannon's Index' (H'), widely employed in ecological literature as Shannon's diversity index, Shannon–Wiener index, or Shannon–Weaver index, was originally introduced by Claude Shannon to quantify the entropy, or information content, in textual strings. Independently derived by Shannon and Wiener in 1963, the Shannon-Wiener diversity index (H') amalgamates equitability and species richness by incorporating the proportion of individuals (p) for each species (i). High values of H' indicate elevated biodiversity. The calculation is expressed through the following equation (Poriya 2015).

$$H' = -\sum_{i=1}^R pi \ln(pi)$$

Where,

H' = Shannon weiner index; R = Species richness (number of species in a community); Pi
= proportion of species i

➤ Simpson diversity Index

Simpson's Diversity Index (D) assesses the likelihood that two randomly chosen individuals from a sample will be of the same species (or another specified category). This index reflects both the variety of species present and the population size of each species. A higher D value indicates a diverse and species-rich habitat, which is considered beneficial, while a lower D value suggests a habitat with limited species diversity, which is viewed as less favourable. It was calculated using following equation,

$$D = 1 - \sum_{i=1}^s pi^2$$

Where,

D = diversity index; S = Species richness (number of species in a community); Pi = proportion of species i

➤ **Pielou's evenness**

The evenness was determined through Pielou's evenness index. This index is a measure of equitability, reflecting how evenly individuals are distributed across different species. Pielou (1966) introduced this concept to quantify the uniformity of an ecological community. It was calculated using following equation,

$$J' = \frac{H'}{\ln(S)}$$

➤ **Menhinick Index**

The Menhinick index is used to assess species richness within the studied ecosystem. The index provides a measure of species richness per individual, accounting for the influence of sample size on observed diversity. It was calculated using following equation,

$$D = \frac{S}{\sqrt{N}}$$

Where,

D = Menhinick index; S = number of species observed; N = total number of individuals sampled

➤ **Margalef Index**

The Margalef index is used to evaluate species richness within the studied ecosystem. The index provides a measure of species richness adjusted for sample size, offering insights into the diversity of ecological communities. It was calculated using following equation,

$$DM = \frac{(S-1)}{\ln(N)}$$

Where,

DM = Margalef index; S = number of species observed; N = total number of individuals sampled.

ii) Population Ecology

$$\text{Density} = \frac{\text{Number of species recorded from all the quadrates}}{\text{Total number of Quadrates}}$$

$$\text{Abundance} = \frac{\text{Total number of species recorded}}{\text{Total number of Quadrates where the species recorded}}$$

$$\text{Frequency (\%)} = \frac{\text{Number of Quadrates where the species recorded} \times 100}{\text{Total number of quadrates}}$$

Statistical Analysis

The monthly data gathered were organized according to seasons, including winter, summer, and monsoon for the seasonal analysis. To evaluate the significance of differences across locations and over time, a one-way ANOVA was employed. Various

statistical methods were applied to assess the overall reliability of the data, following the approach suggested by Sokal and Rohlf in 1969,