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Distribution, Abundance, and Habitat Ecology of Heterobranchs in The Intertidal Region of South Andaman

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Abstract

The present study focuses on the distribution, diversity, and habitat utilization of Heterobranchs in the intertidal zone of the South Andaman Islands from January to March 2017. Samples were collected by the "quadrant method" at three stations (St. 1-Burmanallah (BR), St. 2 - Carbyn's Cove (CR), and St. 3 - Science Centre (SC) of South Andaman. The total number of individuals recorded was 361 belonging to 3 orders namely Cephalispidea (2 species), Sacoglossa (3 species), and Nudibranchia (9 species). A total of 14 species were identified during the study period. Plakobrachus ocellatus was the most abundant species recorded from all three stations and showed high abundance in St. 2 (196 individuals), followed by St. 1 (111 individuals) and St. 3 (10) individuals. The present observation of colour polymorphism in Dendrodoris fumata includes the presence of two distinct forms: an orange-to-red form and a grey form. The study reveals that the population sharing similar habitats have different degrees of distribution and abundance, due to the influence of environmental factors and, the availability of food sources in that habitat. The current study contributes valuable information on Heterobranchs, enhancing the knowledge base for future research in the field of molluscan studies.

Key words: *Heterobranchia, intertidal zone, diversity, polymorphism, South Andaman*

Introduction

The Andaman and Nicobar Islands are the largest archipelago system in the Bay of Bengal, consisting of 836 islands, islets, and rocky outcrops. The total geographical area is 8249 km^2 with a coastline of 1962 km^2 (Government of India, Planning Commission, 2007). The continental shelf area is very limited with an estimated area of 16,000 km^2. The Exclusive Economic Zone (EEZ) around the islands encompasses around 0.6 million km^2, which is again around 30% of the EEZ of India (Fisheries Department, Andaman and Nicobar Administration, 2018). This provides a great opportunity to explore the vast diversity of the seas around these islands.

The first available report of Heterobranchs fauna from India was made by Alder and Hancock (1864). Heterobranchs are one of the less-studied groups of molluscs in the Andaman and Nicobar Islands (Sreeraj et al., 2012). The term Heterobranchia branch means "different gilled snails" which is specific to this group. Studies on this cryptic creature are very less because of its low numerical density and many of it is sub-tidal. Around 3500 species are described worldwide now (Ramakrishna et al., 2010). Heterobranchs possess a radular feeding organ and external gills and mantle cavity typical of other molluscs and are mostly carnivorous, but some are herbivorous and few show cannibalistic feeding habits. Some are referred to as solar-powered" where organism derives their food from the by-products of photosynthetic algae stored in their body (Gosliner et al., 2008).

The molluscan diversity studies of Andaman and Nicobar Islands were started in the late 19th century. The literature available shows that the earliest molluscan study was on a collection of marine shells made by Smith in 1878. The first report on nudibranchs from these Islands was published by Eliot (1910). At the turn of the 2lst century, 29 species of Heterobranchs were reported from Andaman and Nicobar Islands belonging to 11 families and 4 orders, including the occurrence of nudibranch, *Pseudo vermissolcatus* from these Islands (Subba Rao and Dey, 2000). Subba Rao (2000) reported the occurrence of eight orders and as many as fifty families

consisting of about 150 species which is considered a poor representation of the richly diversified sea slugs in India compared to 3400 species estimated to occur in the Indo-West Pacific Province. Heterobranchs taxonomy and ecology in these Islands recently gained attention after 99 years of the publication by Eliot with the report of five species of nudibranchs (Raghunathan et al., 2010) and 17 new distributional records of nudibranchs by Sreeraj et al., 2009 including new records to the Indian Subcontinent. Other works in Andaman and Nicobar Island were carried out by Apte et al., 2015, Sreeraj et al., 2012 a, b, 2013), Baskaran et al., 2013, Sumanata et al., 2013, Shaktivel et al., 2014 and Kiruba et al., 2014 and Mythri, 2015.

Heterobranchs have been popularised due to their commercial trade value in the aquaculture industry and the pharmaceutical potential of their secondary metabolites. (Gosliner et al.,2008). Quantitative studies on opisthobranch populations in Indian waters are scarce (Sreeraj et al., 2012). These species are considered indicator species and are also used as model organisms for advanced scientific research (Wagele et al., 2014). Due to the remoteness of the Andaman and Nicobar group of islands Heterobranchs of these waters are least explored when compared to other world seas, which indicates the importance and significance of the taxonomic study for this unique group. Many Heterobranchs of Andaman waters are threatened species; they require the attention of the scientific community for conserving both habitats as well as the species itself. The present study deals with the distribution, abundance, and habitat ecology of Heterobranchs from selected intertidal sites of South Andaman. This study will be an asset to the taxonomic studies on Heterobranchs from the island ecosystem.

Materials and Methods

Study Area

The study was carried out for a period of 3 months from January 2017 to March 2017. Three stations Burmanallah (BR) Carbyns'Cove (CC) & Science center (SC) of South Andaman were chosen for sampling, based on the pilot survey conducted in December 2016 **(Fig.1).** Stations were selected on the availability of food sources, habitat features, and accessibility.

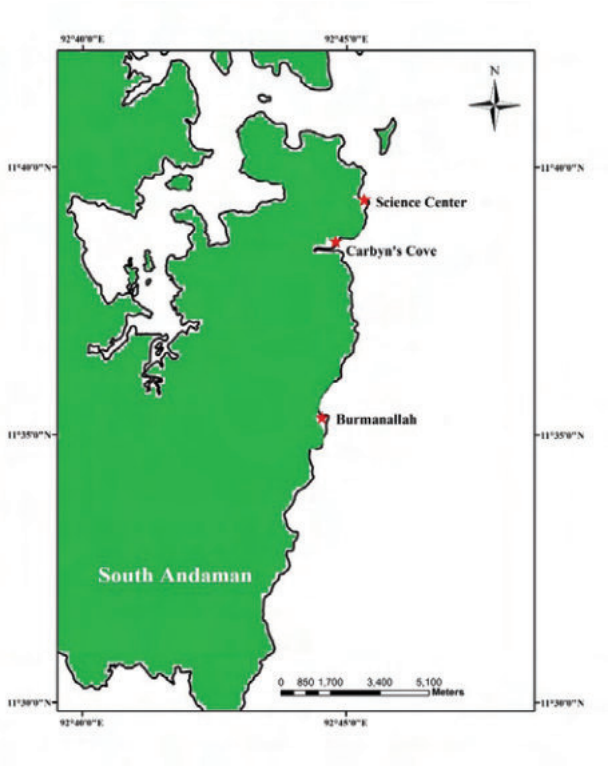


Fig.1. Map showing sampling stations. Inset: The study site is indicated by stars.

Station 1: Burmanallah (BR)

The sampling site is located between 11º333.255' N, 092º43.892' E. Burmanallah has a long rocky shore stretch with mangroves on one side with great biodiversity. The population was collected randomly and assessed by using the quadrant method. Sea slugs, brittle stars, flatworms, crabs, sea anemones, octopus, corals, sponges, seagrass, and seaweeds are abundantly observed in the intertidal zone of this area.

Station 2: Carbyn's Cove (CR)

The sampling site is located between 11º38.428' N, 92º44.652' E. Carbyn's Cove has a long stretch of rocky shore with several tide pools. Brittle stars, mud skippers, damselfish, sponges, and polychaetes were common organisms observed during the study period. The area is relatively polluted due to human settlement and interference.

Station 3: Science Center (SC)

Science Center is an area with huge rocks. Tide pools are comparatively low compared to the other two stations. Common organisms observed during samplings were algae, seaweeds, brittle stars, crabs, and fishes

Sample Collection

The sampling method was biased and based on the availability of the specimens in the intertidal zone during low tide. The aid of snorkeling was also undertaken for the collection of samples in the observed areas. Species abundance and diversity were studied at each station. Species abundance was undertaken by using a 1m^2 quadrant. Most of the species were photographed and measured in their natural habitat and were collected in plastic bottles. The animals were then brought to the laboratory for further taxonomic identification. Specimens were preserved in a 5% formaldehyde solution. Identification was done only based on external morphology and also following the literature "Indospecific sea slugs- A field guide to the world's most diverse fauna" by Gosliner et al. (2008), Sreeraj et al., 2012 and two web-based portals namely The Australian Museum's Sea Slug Forum (http://www.seaslugforum.net) and Nudi Pixel (www.nudipixel.net) Taxonomic changes published recently are incorporated.

Environmental Parameters

Water samples were collected from January 17 to March'17 from St.1, St. 2 & St. 3 to estimate Temperature, Salinity, pH, and Dissolved Oxygen.

1. **Water temperature**: Water temperature was recorded with the help of a Hg- thermometer with ±0.1 accuracy. The temperature was expressed in ° C.
2. **Salinity**: Salinity was measured by placing a drop of water sample over the glass in the refract meter (ATAGO) and the reading was measured in PSU.
3. **pH**: The pH was measured by immersing the pH meter (GENEI) in the water sample and the reading was noted.
4. **Dissolved Oxygen**: Dissolved Oxygen of the water sample was estimated using Winkler's method and the value was recorded in mg/L.

Statistical analysis

All data collected were fed for statistical analysis. Univariate and multivariate analyses were performed using the statistical software PRIMER (version 5). The most commonly used univariate biodiversity indices are d' (Marglef'sspecies richness,1968), J' (Pielou'sevenness, 1966), and H' diversity index (Shannon-Weiner, 1963). These indices were used to calculate the species diversity of the area. Multivariate analyses such as Bray-Curtis similarity and Pearson correlation were performed based on species abundance data to understand the cluster and correlation between stations (BB, CC, and SC) and Canonical correspondence analysis (CCA) was performed to understand the relationship between the species and environmental parameters (temp, salinity, DO and pH) among the three stations.

Results and Discussion

Species Composition

In the present study, a total of 14 species were observed **(Table 1).** The species composition at CC was found to be higher with 8 species followed by BR with 7 species and SC with 3 species.

Table 1. Occurrence of Heterobranchs species in St. 1, St. 2 & St. 3 during the study period

Sl No.	Species Occurred	BR			CC			SC		
		Jan	Feb	Mar	Jan	Feb	Mar	Jan	Feb	Mar
1	*Ceberilla annulata*	-	+	-	+	-	-	-	-	-
2	*Plakobrachus ocellatus*	+	+	+	+	+	+	+	+	+
3	*Phyllidiella zeylanica*	-	+	-	-	-	-	-	-	-
4	*Haminoea ovalis*	-	-	-	-	-	-	+	+	-
5	*Thuridiella coerula*	-	-	-	+	-	+	-	-	-
6	*Phanerophthalmus smaragdinus*	-	-	-	+	+	+	-	-	-
7	*Gymnodoris striata*	+	+	+	-	-	-	-	-	-
8	*G. citrina*	-	-	-	+	-	-	-	-	-
9	*Dendrodoris nigra*	-	-	-	+	+	+	-	-	-
10	*Dendrodoris fumata*	-	+	+	+	-	-	-	-	-
11	*Bornella stellifer*	+	-	-	-	-	-	-	-	-
12	*Phyllidia varicosa*	-	-	-	_	-	-	-	+	+
13	*Chromodoris sp.*	-	-	-	+	-	-	-	-	-
14	*Elysia sp.*	+	+	+	-	-	-	-	-	-

At CC, 8 species belonging to 3 orders were found. Order Cephalispidea was represented by the family Haminiodie in which one species *Phanerophthalmus smaragdinus* was recorded in this study. Order Sacoglossa was represented by the family Plakobranchidae from which two species *Plakobrachus ocellatus & Thuridiella coerula* belonging to the genus *Plakobrachus* and *Thuridilla* were found. Order Nudibranchia showed a greater number of species, represented by the family Dendrodoridiidae, Gymnodoridiidae, & Aeolidiidae. From the family Dendrodoridiidae two species *Dendrodoris nigra & Dendrodoris fumata* belonging to the same genus *Dendrodoris* were found. From the family Gymnodoridiidae, one species *Gymnodoris citrina* belonging to the Genus *Gymnodoris* was found. One species representing the Genus *Cereberilla, Cereberilla annulata* of the family Aeolidiidae was also recorded.

At BR 7 species belonging to 2 orders were found. Order Sacoglossa was represented by the family Elysiidae and Plakobranchidae from which one species, *Elysiida sp*., and *Plakobrachus ocellatus* were found. Order Nudibranchia was represented by the family Phyllididae, Gymnodoridiidae, Dendrodoridiidae, Bornellididae & Aeolidiidae from which a total of 7 species such as *Phyllidiellazeylanica*, *Gymnodorisstriata, Dendrodorisfumata, Bornella stellifer,* and *Cereberilla annulata* each were encountered.

At SC only 2 species belonging to 2 orders Nudibranchia and Cephalispidiea. *Phyllida varicosa* and *Haminoea ovalis* from the family Phyllidiella and Haminioeoidea were observed.

Species Description:

1. *Phyllidiella zeylanica* (Kelaart, 1858)
Spotted site- BR
Distribution- Gulf of Kutch (Apte et al., 2010)
Preferred habitat- Rock bed and corals
Size- 2 cm
Description- The dorsal surface of the mantle has five longitudinal rows of pink tubercles which are arranged in a semicircular fashion along the anterior and posterior margins except the median one. Irregular tubercles are arranged in a group of two or three. The median ridge forms a canal-like structure at the centre which is filled in by black lines. These lines, except the median one is all along the posterior and anterior margins in a semicircular fashion (Narayanan, 1969). Rhinophores are black with colourless stalks. The foot stole is white.

2.*Phyllidia varicosa* (Lamarck, 1801)
Spotted site- SC at 2m depth
Distribution- Throughout the Indo-West Pacific Ocean including the Red Sea, India- Andaman & Nicobar, and Gulf of Mannar (Apte et al., 201, Sreeraj et al., 2012)
Preferred habitat- Rock surfaces and corals
Size- 4.5cm
Description- The most common nudibranchs are distributed in the Andaman Islands. The length of the collected specimens ranged from 3.5 cm - 9.7 cm and the width varied from 1.3 cm - 3.3 cm. The body is firm, 3-6 longitudinal tuberculate, not all bluish-grey ridges on the dorsum. The ridge and bases of the tubercles are blue-grey and the tubercles are capped in yellow. The foot sole possesses a black longitudinal foot stripe. The Rhinophoral clavus possesses 27-30 lamellae.

3.*Gymnodoris citrina* (Bergh, 1877)
Spotted site- CC
Distribution-Wide spread in Indo-West Pacific Ocean, India (Laccadives, Andaman), and Australia (Apte et al., 2010, Sreeraj et al., 2009)
Preferred habitat- Found on rock surfaces along with corals
Size- 2.5 cm
Description- The translucent white body has many oranges to yellow spots. The gills are white in colour. The white rhinophores are tipped with yellow colour and the rhinophoral lamellae are white. Anterior margin of the head with a distinctive row of tubercles. Gill branched from a complete circle. The anterior ends of the foot end in two-lobed sensory folds.

4.*Gymnodoris striata* (Eliot, 1908)
Spotted site- BR
Distribution- Indo-West Pacific, from Red Sea to Japan, India: Ritchies Archipelago, South Andaman (Sreeraj et al., 2009, Ramakrishna et al., 2010)
Preferred habitat - Found along with Seagrass. Feeds on *Plakobrachus ocellatus*
Size- 4.5 cm
Description- It is characterized by the longitudinal orange lines on the dorsum and the transverse arrangement of the gills. There is considerable variability in the orange ridges on the body. The upper half of the rhinophore is also yellowish orange.

5. *Cerberilla annulata* (Quoy and Galmard, 1832)
Spotted site- CC& BR
Distribution- Indo-West Pacific distribution.India: Mayabunder (North Andaman) (Sreeraj et al 2012, Ramakrishna et al (2010)
Preferred habitat -Intertidal to shallow, sandy slopes, usually active at night.
Size- 3-4 cm
Description- Mantle and cerata are white. This sand-dwelling aeolid has a wide tropical Indo-West Pacific distribution. It's characterized by the relatively long cerata, white body and cerata, and yellow (upper) and black (lower) bands near the central tip. The yellow and black bands are divided into small white bands. The cerata are numerous and are arranged irregularly.

6.*Bornella stellifer (*Adams & Reeve 1848)
Spotted site- BR
Distribution- India (Gulf of Kutch, Gulf of Mannar), Australia, South Africa (Apte et al., 2010)
Preferred habitat - Rocks where hydroids are present
Size- 3-4 cm
Description- A small sea slug found on rocky reefs. Oral tentacles are paired and finger-like. Gills are placed at the base of each cerata. Rhinophores are present on long stalks & surrounded by long papillae. Deep reddish brown with white patches. Tip of cerata and papillae with apical red band.

7. *Dendrodoris nigra* (Stimpson 1855)
Spotted site- CC
Distribution- South Africa, Madagascar, Indonesia, Hawaii, and Andaman & Nicobar Islands (Sreeraj et al., 2012, Ramakrishna et al.,2010)
Preferred habitat – Around corals and sponges
Size: 4 cm
Description- As the name implies the organism is black in colour. Have clusters of white spots on the mantle. Pease (1871) also gave different names *(D. debilis/ D. sordida/ D. rubrilineata),* to colour forms of this species. Most external differences between *D. nigra* and *D. fumata* are comparative. For example, *D. nigra* is elongate when crawling more than *D. fumata.* The position and nature of the gills would appear to be characteristic. In *D. nigra* the gills are relatively numerous and form a compact

circle around the anusat, the extreme posterior end of the dorsum. The rhinophoral tip is white in colour. The white spots on the mantle are prominent in the center.

8. *Dendrodoris fumata (*Ruppell & Leuckart, 1830)
Spotted site- CC and BR
Distribution- South Africa, Madagascar, Japan, Tanzania, Japan, Andaman (Sreeraj et al., 2012, Ramakrishna et al., 2010)
Preferred habitat – Around sponges and corals in shallow water
Size-1.6 cm
Description- It differs from *D.nigra* in having fewer large gill branches which spread &extend to the edge of the mantle. Species show colour variations from brown to red.

9. *Hamenia ovalis* (Pease, 1865)
Spotted site- SC
Distribution- West coast of India, Andaman & Nicobar, Australia, Samoa, Japan, Guam. (Sreeraj et al., 2012, Ramakrishna et al., 2010)
Preferred habitat- Herbivorous, found feeding exclusively on filamentous algae in intertidal rock pools.
Size- 1.5 cm
Description-Shell is fragile and transparent. The organism is brilliantly colored. The light green color is profusely spotted with orange spots which are encircled by light green and also bears deep blue spots on the mantle and foot. The Foot is short spotted orange and blue.

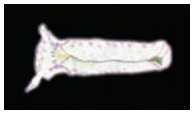
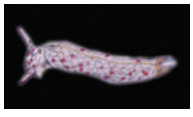
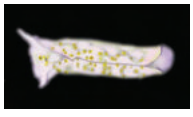
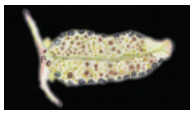
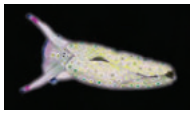
10.*Phanerophthalmus smaragdinus* (Ruppell & Leuckart, 1828)
Spotted site- CC
Distribution- Western Indian Ocean South Africa, Andaman & Nicobar, Madagascar (Sreeraj et al., 2012, Ramakrishna et al., 2010)
Preferred habitat- Found in the intertidal area, Crawling through algae over rocks
Size- 1.8 cm
Description-The animal is long and cylindrical with a very small head shield with two eye spots. The mantle cavity was much reduced and occupied a terminal position at the end of the body. The shell was small, flattened, and enclosed by the mantle. The sides of the foot extended into large parapodia which folded over and enclosed the body (Rudman, 1972). The uniform light greenish colour of this species is distinctive.

11. *Thuridilla coerulea* (Kelaart, 1858)
Spotted site- CC
Distribution- Andaman & Nicobar Islands. (Sreeraj et al., 2012, Ramakrishna et al., 2010)
Preferred habitat- Intertidal muddy to rocky shore with algae and other seaweeds.
Size- 1.6 cm
Description- *Thuridilla coerulea* is blue with red-orange and black markings. The oral tentacles are orange-red with a black edging and the upper half of the rhinophores are black with a central black band. The parapodia are blue with a broad red-orange margin and an inner thin black line. Wings are folded. Tentacles two, blue with a central red ring and blackish tip.

12.*Plakobranchus ocellatus* (Van & Hasselt, 1824)
Spotted site-CC and BR
Distribution: Widespread throughout the tropical Indo-West region (Sreeraj et al., 2012, Ramakrishna et al., 2010, Apte et al., 2010)
Preferred habit- Rocky bed covered with algal patches.
Size- 2-2.3 cm
Description- *Plakobranchus ocellatus* is a common inhabitant of shallow waters of the Andaman Islands. This species was reported in India four decades ago (Vrabhadra Rao, 1961). Although several names exist for various color forms, the consensus is that there is only one species. **Table. 2.** It is mostly seen half buried in the sandy area or coral reef of intertidal areas like species of *Elysia,* it has parapodia that fold over the backside of the animal. The green patch on the mantle is from the green ridges, packed full of microscopic chloroplasts, which line the inside of the parapodia. The rhinophore is creamy white.

13. *Elysia* sp.
Spotted site – BR
Distribution- Andaman & Nicobar Islands (Sreeraj et al., 2012, Ramakrishna et al., 2010)
Preferred Habitat- Rhodophyta
Size- 1.3 cm
Description- Parapodial flaps lie perpendicular to the body. They meet and point upwards even when folded totally

Table 2. Distribution, external morphology, and colour pattern of *Plakobranchus* varieties identified as *P. ocellatus* and cite the reference.

Taxon	Color pattern	Distribution
P. ocellatus var. A 	Small ocellate spots and white spots scattered over the head and dorsal flaps. Transverse dark stripes. Foot sole with some white and dark spots and some scattered ocellate spots.	Okinawa, Japan; Bai Su, Vietnam; Milne Bay and New Hanover, Papua New Guinea; Cebu, Philippines; Ambon, Indonesia
P. ocellatus var. B 	Body white cream, ocellate spots with red-brown rings present over the head and lateral flanks. Dorsal white spots. Foot sole with black spots.	Okinawa, Japan
P. ocellatus var. C 	Translucent white rose body with few dark red ocellate spots over the parapodia and head. Foot sole with small dark dots green and brown.	Okinawa, Japan; Mayotte
P. ocellatus var. D 	Dorsal body white-green, scattered with yellow spots. Foot sole white green, without spots.	Okinawa, Japan
P. ocellatus var. E 	Translucent olive-green body, covered with large white- cream spots, rhinophores, oral tentacles, and tail with bluish-purple tips. The tail tip is also black. Foot sole blue.	West Papua and Sulawesi, Indonesia; Cebu, Philippines; Palau; Milne Bay and New Hanover, Papua New Guinea; Nha Trang, Vietnam; Okinawa, Japan
P. ocellatus var. F 	Rhinophores, oral tentacles, and tail with black tips. Head and parapodia cover with many ocellate spots, small yellow and medium dark. Lateral flanks with largest black ocelli. Foot translucent green with few black ocelli.	Okinawa and Kagoshima, Japan
P. ocellatus var. G 	Body rose-brown, with white spots and some scattered brown and blue spots.	Moorea, French Polynesia
P. ocellatus var. H 	Body white-light green with a little rose on the head. Yellow ocellate spots all over the parapodia, only two on the head. Lateral flanks with large green ocellate spots. Oral tentacles and rhinophores tips blue. Rhinophore with a fuchsia sub-terminal ring.	Cebu, Philippines

14: Unidentified juvenile in this study
Spotted site- CC
Preferred habitat- under rock crevices
Size- 1.4 cm
Description- Slender white body with 2 spots - violet and red at the anterior portion. Also, has a long antenna.

Abiotic Variables

Environmental parameters such as temperature (°C), salinity (PSU), pH, and dissolved oxygen (mg/L) for Burmanallah (St.1), Corbyn's Cove (St.2), and Science Center (St.3) during the study period from Jan'17 – Mar'17. **Table. 3**

Table 3. Environmental parameters recorded at BR, CC, and SC during the study period

Parameters	BRJ	BRF	BRM	CCJ	CCF	CCM	SCJ	SCF	SCM
Air Temperature (°C)	30	26	28	30	26	28	30	32	30
Water Temperature (°C)	33	28	29	33	28	29	28	34	29
DO (mg.L^{-1})	3.3	5.7	4.9	3.3	5.7	4.9	4.2	4.5	3.5
Salinity (PSU)	32	33	30	32	33	30	31	35	28
pH	7.9	8.3	8	7.9	8.3	8	7.7	8.1	7.5

The highest air temperature was recorded at SC (32°C) in February and the least (26 °C) at BN in the same month. The highest water temperature was noted at CC (36°C) in March and the least at BN & SC (28°C) during January and February respectively DO was high at CC (6.8mg/L) in March and least at BN(3.3mg/L) in January. Maximum salinity was recorded at SC & CC (35 PSU) during January and February and the least at SC (28) in March. pH was maximum at CC (8.5) in February and minimum at SC (7.5) in March (**Fig .2, Fig.3, Fig. 4)**

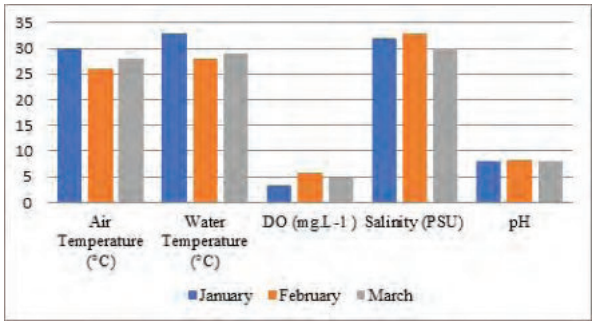


Fig.2. Physico-chemical parameters recorded at Burmanallah from Jan'17 to Mar'17

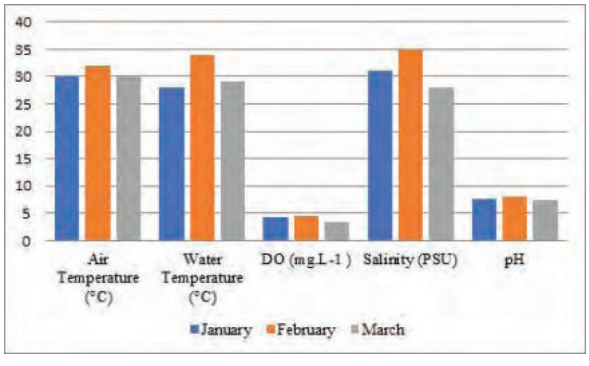


Fig.3. Physico-chemical parameters recorded at Carbyn's Cove from Jan'17 to Mar'17

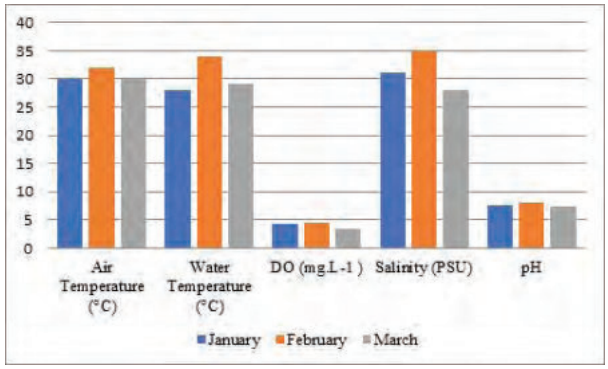


Fig.4. Physico-chemical parameters recorded at the Science center from Jan'17 to Mar'17

Multivariate analysis

Cluster analysis (Bray Curtis similarity index) showed two distinct clusters based on stations. Station 1 (BR) and Station 2 (CC) formed one cluster due to the high abundance of *Plakobranchus ocellatus* and *Cerberilla annulata* while Station 3 (SC) out layered due to the absence of most of the species found in St.1 and St. 2 **(Fig. 5).** Upon performing nMDS it could not differentiate between the two stations namely CC and BR. To verify the cluster between St.1 and St.2., PCO (Pearson Correlation) was performed where segregation was observed between the stations signifying the higher abundance of *Plakobranchus ocellatus* in St.2 **(Fig. 6).**

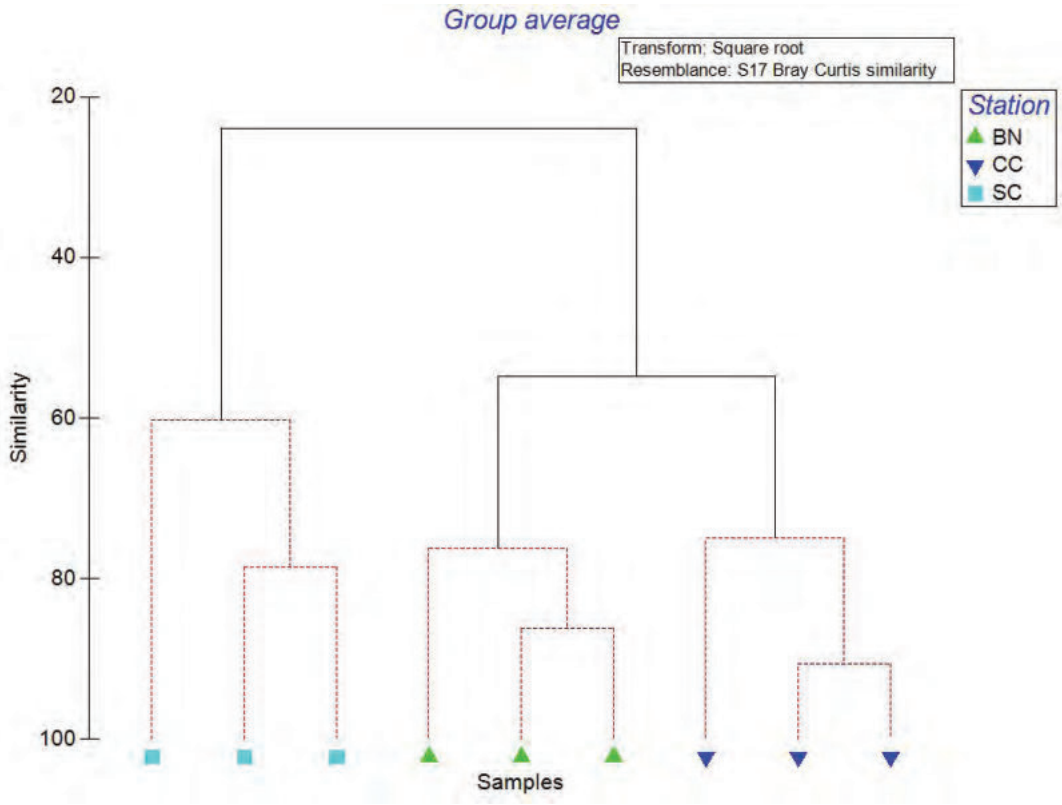


Fig.5.Bray-Curtis similarity shows the formation of clusters between stations in the study area.

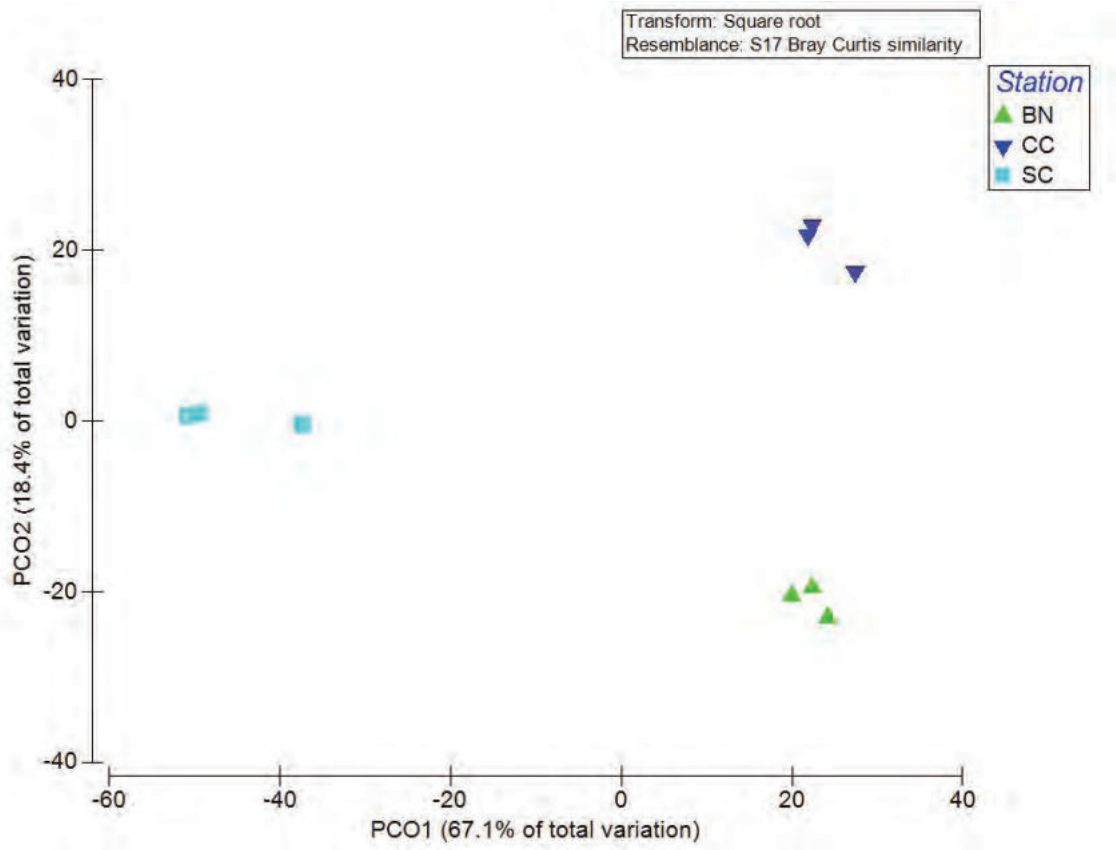


Fig. 6. Pearson Correlation showing segregation between St.1 and St.2

Canonical correspondence analysis (CCA) was performed to elucidate the relationships between Heterobranchs species and the environmental parameters among the three stations. In the ordination diagram (**Fig.7**); the species score, the site scores, and the environmental scores are plotted in the figure- Triplot. Along the axes, during the study period, the high abundance of *Plakobranchus ocellatus* and *Cerberilla annulata* showed a positive correlation with dissolved oxygen and pH. Species such as *Dendrodoris fumata*, *Dendrodoris nigra*, *Thuridiella coerula*, *Gymnodoris citrina*, *Chromodoris* sp. showed a positive correlation with salinity while species such as *Elysia* sp., *Gymnodoris striata,* and *Bornella stellifer* showed a positive correlation with water temperature. One-way ANOVA, Mann-Whitney, and Kruskal Wallis tests were performed but no significant difference was observed.

Species diversity

At St. 1, the maximum no. of species (S = 6) was recorded in February where relatively higher species diversity (H' = 1.235) and species evenness (J = 0.48) was observed. At St. 2, the maximum no. of species (S = 8) with high species diversity (H' = 1.0) and Margalef's richness (d = 1.7) was recorded during January which could be due to the dominance of few species such as *Plakobranchus ocellatus* and *Cereberilla annulata* which showed low equitability in the population (J'=0.37). At St. 3, the maximum no. of species (S = 3) was recorded during February where relatively higher species diversity (H' = 1.37) and high species evenness (J = 0.9) was observed **(Table.2)**

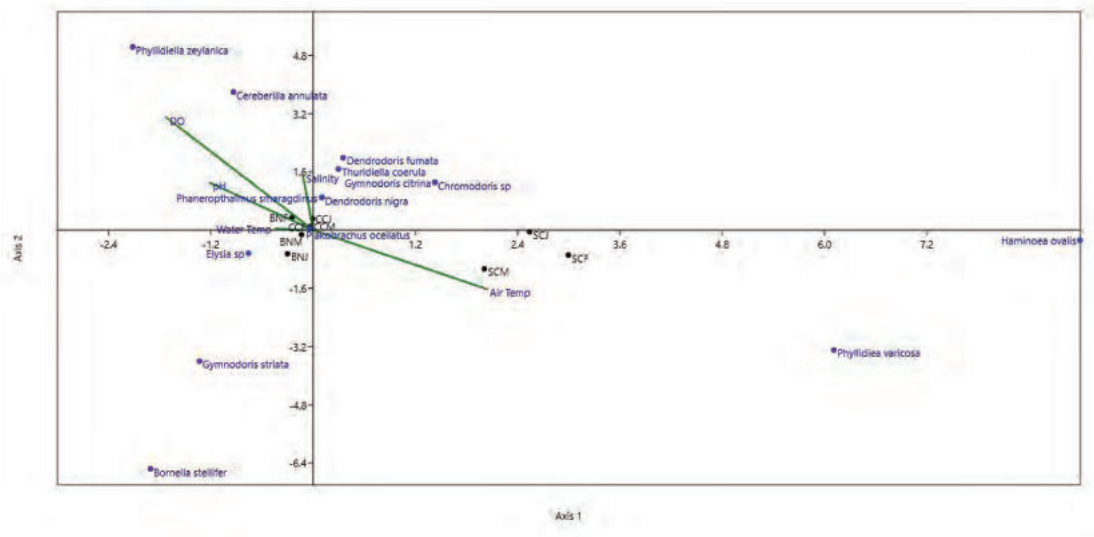


Fig.7. Canonical correspondence analysis (CCA) was performed to elucidate the relationships between Heterobranchs species and the environmental parameters among the three stations.

Table 4. Diversity indices of Heterobranchs in the study area

Stations	Number of Species (S)	No of individuals (d)	Marglef's species richness (d)	J'-Pielou's evenness (J')	Shannon-Weiner diversity index) H'(log2)
BRJ	4	53	0.755612	0.482099	0.964197
BRF	6	45	1.313487	0.478051	1.235744
BRM	4	39	0.818875	0.373807	0.747614
CCJ	8	58	1.72395	0.319368	0.958105
CCF	3	84	0.451384	0.117439	0.186136
CCM	4	67	0.713489	0.167373	0.334746
SCJ	2	7	0.513898	0.863121	0.863121
SCF	3	5	1.24267	0.864974	1.370951
SCM	2	3	0.910239	0.918296	0.918296

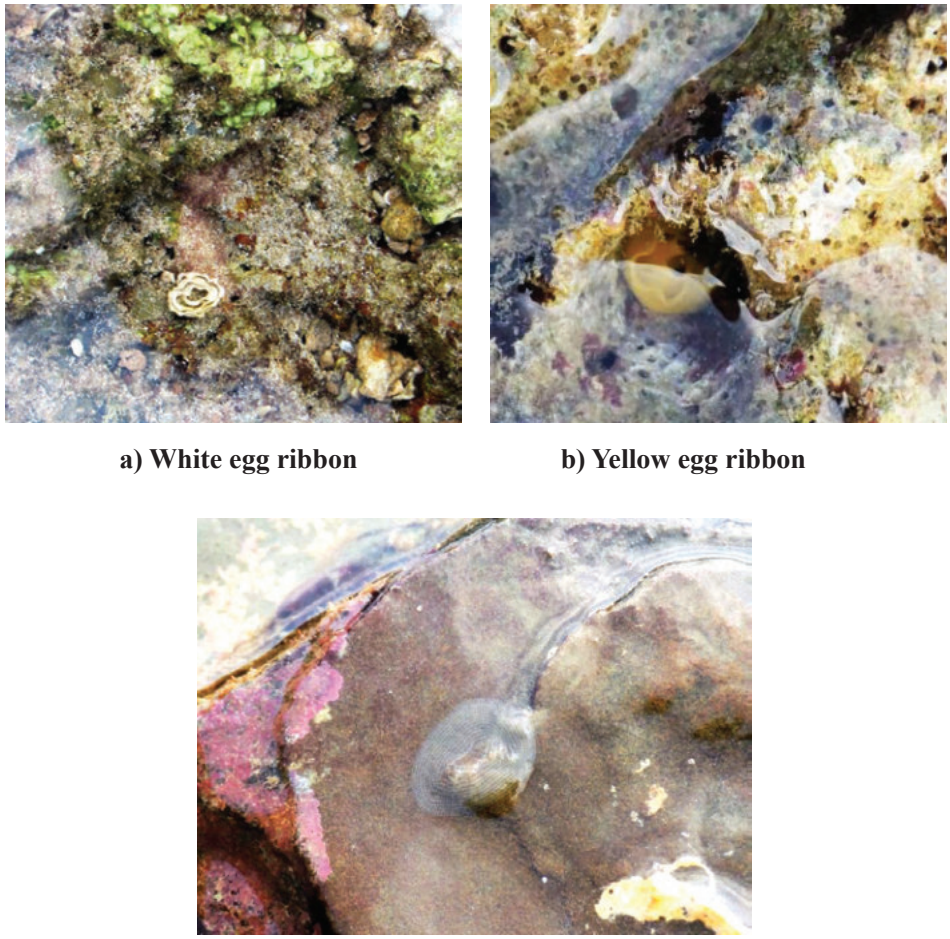
*BN - Burmanallah, CC-Carbyn's Cove, SC-Science Center, J-January, F-February, M-March

The egg masses

Three spiral-shaped egg masses one from St 1 and two from St 2 were observed during the study period **(Fig.8 a-c).** The white ribbon-shaped egg masses were observed from St 1 which is of Goniodoris sp. The yellow ribbon-shaped and white crystal-shaped egg masses were observed from St 2 which is of Chromodoris species and Elysia sp respectively.

In the present study, investigated Heterobranchs population was represented by 361 individuals; 3 specific orders and a total of 14 species were confined to these orders **(Fig .9).** Nudibranchia was the most common

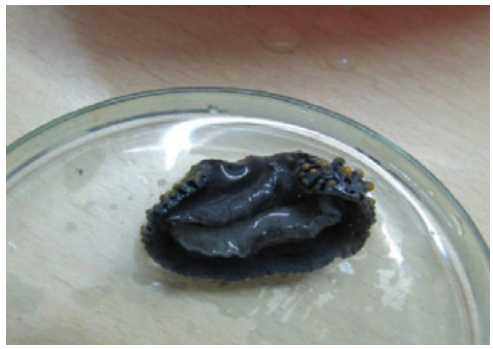
order found in the SC (St. 3) and BR (St. 1) and CC (St. 2). The order Nudibranchia showed higher diversity and abundance in St. 1, even though St. 2 showed higher diversity and abundance of whole Heterobranchian population. Higher Nudibranch diversity from St. 2 is due to the availability of optimal abiotic conditions as found in this study has been reported earlier from this area (Mythri, 2015). Individuals belonging to Cephalispidea were recorded from St. 3 and St. 2, and members of the order Sacoglossa were recorded from St. 1 and St. 2, All these three orders were recorded from St. 2, the higher degree of diversity found in St. 2 is due to the availability of rich food source, lesser competition in the community & scarcity of predatory influence in this area. Stations such as St. 1 and St. 2 were the active breeding grounds of Heterobranchs, egg masses were abundantly observed during January, and juveniles were observed during February, indicating these are the periods to spawn as successful breeding was found for this group. The results coincide with previous work done by Claverie et al., (2008). These months showed sudden variation in water temperature (28°C - 35°C) which act as an important factor promoting the development and hatching of young ones. Both juveniles and egg mass were absent in the Science Center even though this station had all the optimal conditions for the successful development of Heterobranchs. This is due to the presence of carnivorous predators like crabs that were generally observed in this area during collection.



a) White egg ribbon **b) Yellow egg ribbon**

Fig.8. a-c The Egg Masses of Heterobranchs recorded during the study

Phyllidiella varicosa (Dorsal view)

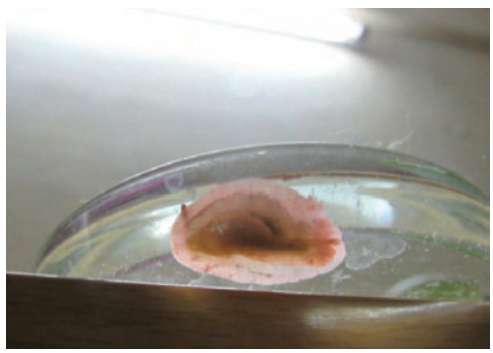


P.varicosa (Ventral view)

Gymnodoris striata (Dorsal view)

G.striata (Ventral view)

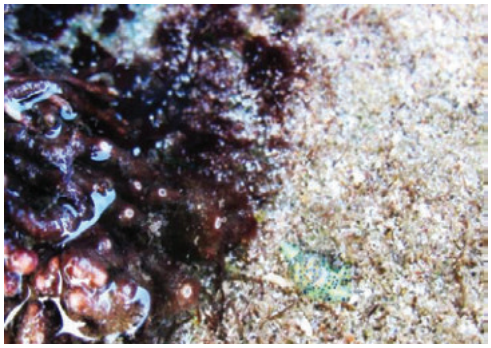
Dendrodoris fumata (Dorsal view)



D. fumata (Ventral view)

Dendrodoris nigra

Cerberilla annulata

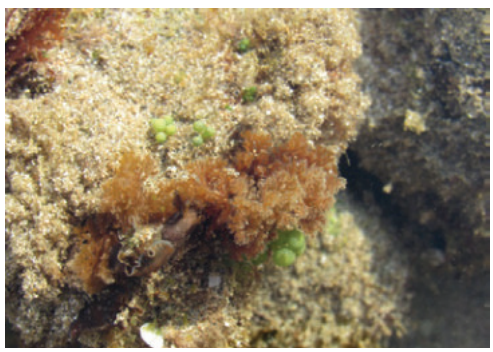


Haminoea.ovalis

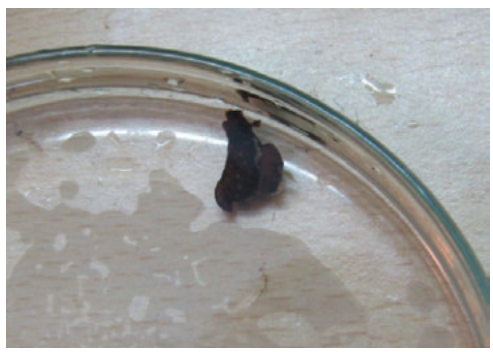
H. ovalis ventral view

Thuridilla coerula

T.coerula (ventral view)



Elysia sp.

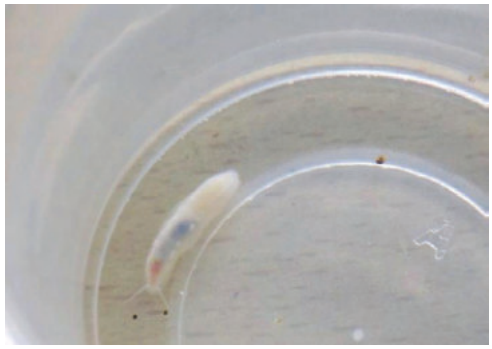


Elysia sp. (Ventral view)

Phyllidiella zeylanica

P. zeylanica (Dorsal view)

Bornella stellifer

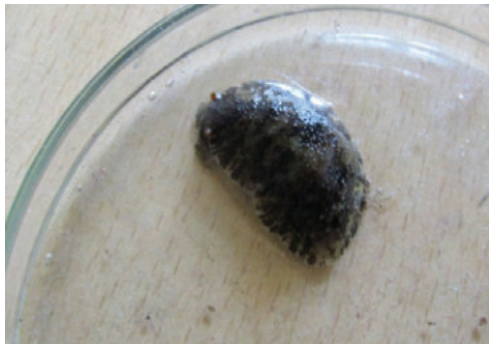


A Chromodoris sp. juvenile

Plakobrachus ocellatus (Orange colour)

Plakobrachus ocellatus (Green colour)

Dendrodoris. fumata
(Orange to red and light pink)



D.fumata (Grey form)

Gymnodoris citrina

Phaneropthalmus smaragdinus

Fig.9. Heterobranchs recorded during the study period

It was reported that *P. zeylanica* is a very rare Nudibranch in India (Burn, 1970) but it is very common in the Andaman Islands (Sreeraj et al., 2010). Color polymorphism of Heterobranchs (Eliot, 1905 and Kay, 1979) was shown by both juveniles and adults of species. Two major polymorphic species recorded during this study were *Dendrodoris nigra*, and D. *fumata*. The adults of *D. nigra* and *D. fumata* showed black colouration, whereas their juveniles were having a yellowish-red color. Species such as *Plakobrachus ocellatus* were found crawling during the evening when the temperature dropped, found in clusters where there were algal patches (*Anaebena* sp) and other macroalgae. *Gymnodoris striata* were also found in between seagrasses patches where there were numerous *Plakobrachus ocellatus* suggesting the carnivorous nature of *Gymnodoris* sp. (Goslinear et. al., 2008). *Plakobrachus ocellatus* is a solar-powered sea slug that absorbs chloroplast from the algae it grazes and stores in the epithelial cells of the digestive gland (kleptoplast). Under starved conditions, they can survive using these kleptoplast photosynthates (Maeda et al., 2012). Three specimens of *Plakobrachus ocellatus* that were collected from the field were found to survive 4 days without food suggesting its utilization of algae as a food source.

The similarity among the species was depicted through the Bray-Curtis similarity index, which showed the formation of two distinct clusters, clustering of Station 1 (BR) and Station 2 (CC) due to the high abundance of *Plakobranchus ocellatus* and *Cerberilla annulata.* The segregation patterns of stations observed in PCO (Pearson Correlation) analysis justified the abundance of the *Plakobranchus ocellatus* and *Cerberilla annulata* throughout the study period. Canonical correspondence analysis (CCA) showed the relationship between environmental parameters and Heterobranchian species, from all three stations. CCA results indicate that multiple environmental factors or a single factor can influence the distribution and abundance of a single species, or different species. *Plakobranchus ocellatus,* the most dominant species recorded during the study showed a correlation with pH and D.O, whereas *Elysia* sp. showed a positive correlation with a single parameter, water temperature. *Dendrodoris niger* always showed a positive relation to DO. Studies correlating environmental parameters with that of Heterobranchs diversity among Indian waters were not available and most of the studies were related to taxonomy. Cyrne et al.,2018 observed the presence of *D. herytra* and *D. grandiflora* in the intertidal area of the west coast of Portugal consistently related to a peak in turbidity and dissolved oxygen. The species which showed correlation with multiple environmental parameters showed higher abundance and a wide range of distribution throughout all three stations, indicating that these species (viz, *Plakobranchus ocellatus)* have acquired high adaptive capability inherited through evolution (Mythri, 2015) Heterobranchs that correlated to a single environmental parameters wiz *Gymnodoris citrina,Gymnodoris striata, Bornella stellifer, Phyllidiea vericosa, Elysia sp,Chromodoris sp., Phanerophthalmus smaragidinus, Thuridiella coerula, Haminoea ovalis, G. Striata, G.citrina, Bornella stellifer* showed a restricted distribution to a particular study area. Both *Thuridiella coerula* and *Plakobranchus ocellatus* belong to similar order *Sacoglossa*, only *Plakobranchus ocellatus* shows a wide distribution all over the three stations, this shows the role of environmental factors on the successful distribution of a particular species from its phylogenic sister species. Temperature variation was found to be the main factor modulating the Heterobrachs diversity (Betti et al.,2017). However similar investigation of intertidal Penghu Island, Taiwan (Su et al., 2009) found no significant variations among the Heterobranchs species count in relation to environmental factors.

Summary and Conclusion

The Present study entitled "Diversity, distribution and habitat utilization of Heterobranchs" was carried out along the three intertidal regions of South Andaman from January 2017 to March 2017. Samples were collected by the "quadrant method" from three distinct areas, St. 1 (BR), St. 2 (CC), and St. 3 (SC). The total number of individuals recorded was 361 belonging to 3 orders namely Cephalispidea (2 species), Sacoglossa (3 species), and Nudibranchia (9 species). A total of 14 species were identified during the study period. The highest species diversity was observed at St. 2 (8 species) followed by St. 1 (7 species) and the least from St. 3 (2

species). *Plakobrachus ocellatus* was the most abundant species recorded from all three stations and showed high abundance in St. 2 (196 individuals), followed by St. 1 (111 individuals) and St. 3 (10) individuals. *Gymnodoris striata* was the second most abundant species with 12 individuals recorded from St. 1. Other species showed a random distribution all over the study area. Bray Curtis analysis showed clustering of St. 1 and St. 2 was due to the wide geographical distribution of the most abundant species *Plakobranchus ocellatus*. Environmental parameters such as temperature, salinity, pH, and DO show a direct correlation with the distribution of all species. Highly abundance of species was influenced by multiple environmental factors which shows their broad range of adaptability. A study on the colour polymorphism of various life stages of opisthobranchs culminated in the finding of variant polymorphic forms of *Dendrodoris fumata.* Colour polymorphism of *Dendrodoris fumata* (orange to red form, and grey form) was recorded. This present study testifies that the Heterobranchs population sharing similar habitats have different degrees of distribution and abundance, it varies according to the influence of environmental factors and the availability of food sources in that habitat.

Three egg masses, 2 from St.2 and 1 from St.1 were also recorded. The presence of Heterobranchs was always found to be along with other organisms such as brittle stars, polychaetes, worms, corals, sponges, etc. diffidently which indicates a healthy ecosystem. (Sabdano et al., 2022). Heterobranchs are rich in secondary metabolites having immense biomedical potential (Fontana, 2006). The present study act as a baseline for further intensive research in the Heterobranchs group and also for molluscan studies in the future.

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