

CHAPTER 4: RESULTS AND DISCUSSION

4.1 DIVERSITY OF SEA ANEMONES

Total 21 species of Sea anemone belonging to 11 genera and 5 families are recorded from the Saurashtra coast of Gujarat. Of which 13 species are first time recorded from Gujarat. Geographical range extension of some of the sea anemone species has been also documented from the study.

Family Actiniidae (Rafinesque, 1815)

Actiniidae with basilar muscles, Column with verrucae, acrorhagi, pseudoacrorhagi or vesicles may be smooth or dotted, but without acontia. Sphincter is either absent or endodermic and can be either diffuse or circumscribed. Tentacles are usually simple, with bifurcated tips at times. Non-divided mesenteries into macro and micro-cnemes, More than six pairs of perfect mesenteries.

Genus *Actinia* (Linnaeus, 1767)

Actiniidae can having wide pedal disc and smooth low column. A ring of simple marginal spherules existing in the deep fosse. Sphincter muscles are weaker or stronger, diffuse, hardly ever with a mild tendency to be meso ectodermal. Tentacles retractile of normal length, their longitudinal muscle tissue ectodermal. Perfect mesenteries are numerous. All stronger mesenteries, keep the directives, fertile. More mesenteries are existing at the base than at the margin. Retractor muscles of the mesenteries are diffused (Carlgren, 1949 and Ottaway, 1975).

1. *Actinia equina* (Linnaeus, 1758) (Plate: 4.1)

Synonyms:

Actinia mesembryanthemum (Johnston, 1847)

Actinia cerasum (Dalyell, 1848)

Actinia corallina (Risso, 1826)

Actinia hemisphaerica (Pennant, 1777)

Actinia margaritifera (Templeton, 1836)

Actinia purpurea (Cuvier, 1798)

Priapus equinus (Linnaeus, 1758)

Priapus ruber (Forsskal, 1775)

Common Name: Beadlet anemone, Red rock anemone

Material Examined: [VAJAE01 (20°49'40''N, 70° 29' 18.6''E) (CH-20mm, CD-12mm, LD- 17mm, PDD-19mm, ODD-20mm), VAJAE02 (20°48'46.26''N, 70°31'12.9''E) (CH-14mm, CD- 9mm, LD-11mm, PDD- 14mm, ODD- 13mm), VAJAE03 (20° 49'39.216''N, 70°29'16.836''E) (CH- 16mm, CD-12mm, LD-13 mm, PDD-15mm, ODD-17mm) ,VAJAE04 (20°48'51.012''N, 70°31'9.84''E) (CH- 11mm, CD-7mm, LD-8 mm, PDD-10mm, ODD-11mm)] collected from the rocky coast of Vadodra Jhala; [SUTAE05 (20°49'35.76''N, 70°22'38''E) (CH-14mm, CD-10mm, LD-11mm, PDD-13mm, ODD-13mm), SUTAE06 (20°50'24''N, 70°27'2''E) (CH-9mm, CD-4mm, LD-5mm, PDD-7mm, ODD-10mm), SUTAE07 (20°48'54''N, 70°30'54''E) (CH-5mm, CD-3mm, LD- 4mm, PDD-5mm, ODD- 6 mm)]collected from the coast of Sutrapada.

Description: These anemones inhabit shaded vertical rock walls and tide pools, usually found in clusters in supra littoral and mid littoral intertidal zones. It is highly adapted to the intertidal zone as it tolerates high temperatures and desiccation. They stick to the rocks only a few feet from the surface or to rocky bottoms that are simply underneath the tide line. Like the entirety of their variety, they feed at high tide and encase themselves into a jelly blob at low tide. The average sized Beadlet anemone is 45 mm. This anemone has a base up to 50 mm in diameter, which is moderately or firmly adhesive. Base formed like a sucker through which it fixes itself to the ground or rock. The tentacles are pointed and smooth with crown of 190 up to 2 cm long, arranged in six circles around its oral disk. The oral disc has not any pattern on it. This Anemone observed in different colours like red, orange and brown. Genetic investigation suggests that some of the different colour morphs of *Actinia equina* may be distinct species (Wilding and Weedall, 2019). Reproduction is sexual as well as asexual with separate sexes or hermaphrodites. Additionally,

Beadlet Anemones can bud to produce asexually new separate individuals. *A. equina* shows aggressive behaviour towards neighbouring individual, which is influenced when the tentacles of adjacent anemones come into contact (Picton and Costello, 1998). The aggressor stings the victim with nematocyst, in the acrorhagi, which leads to the victim both crawling away and dropping off the substratum. Perfect mesenteries are numerous. Mesenteries of Retractor mussels are diffuse (Plate: 4.2).

Distribution: Beadlet Anemones are found in subtropical waters in the Mediterranean and in the colder waters of the Eastern Atlantic Ocean around Britain and Ireland (Kruger and Griffiths, 1996; Nichols and Cooke, 1971). We reported this species from the Sutrapada, Vadodra jhala and Veraval (Gir Somnath District) of the Saurashtra coast, Gujarat.

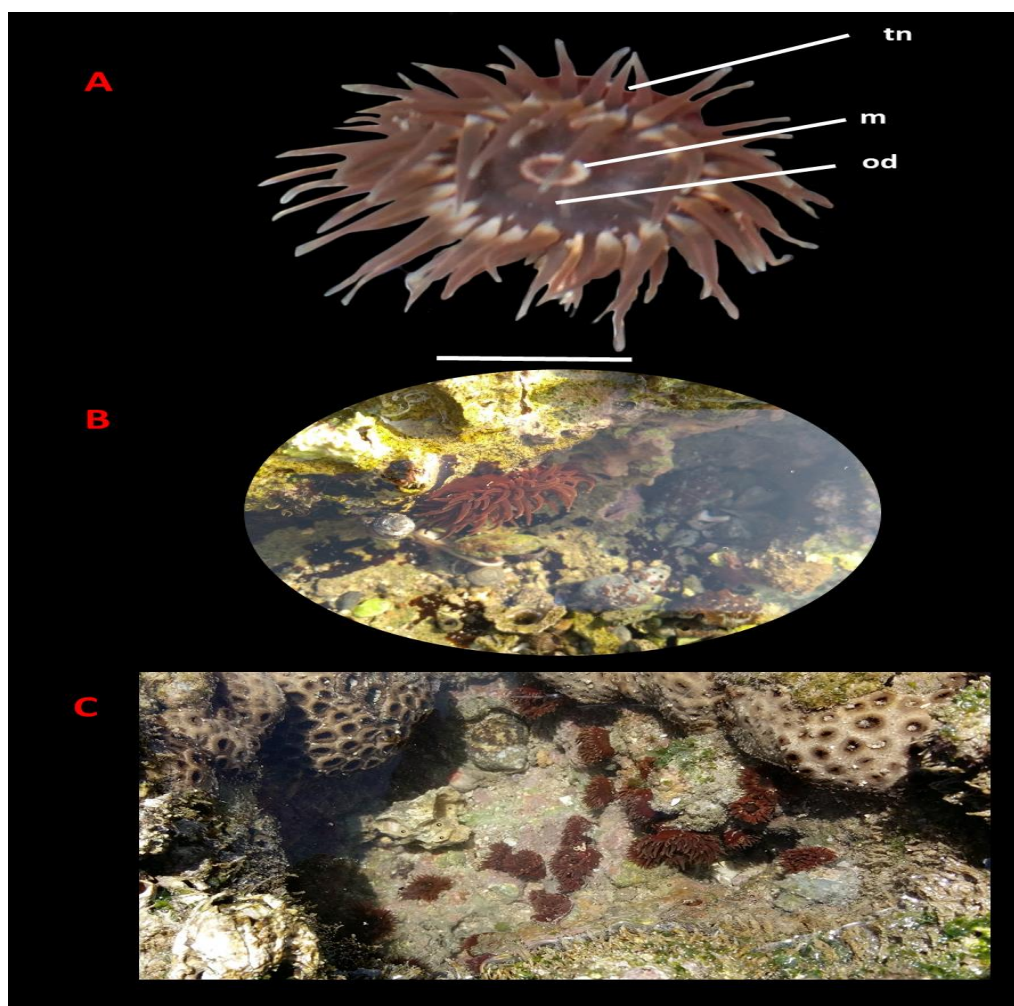


Plate 4.1: *Actinia equina* (Linnaeus, 1758). A: Oral view of living specimen B, C: Specimen in natural habitat. Mouth (m), Oral disc (od), Tentacles (tn).

Scale bar: 10mm

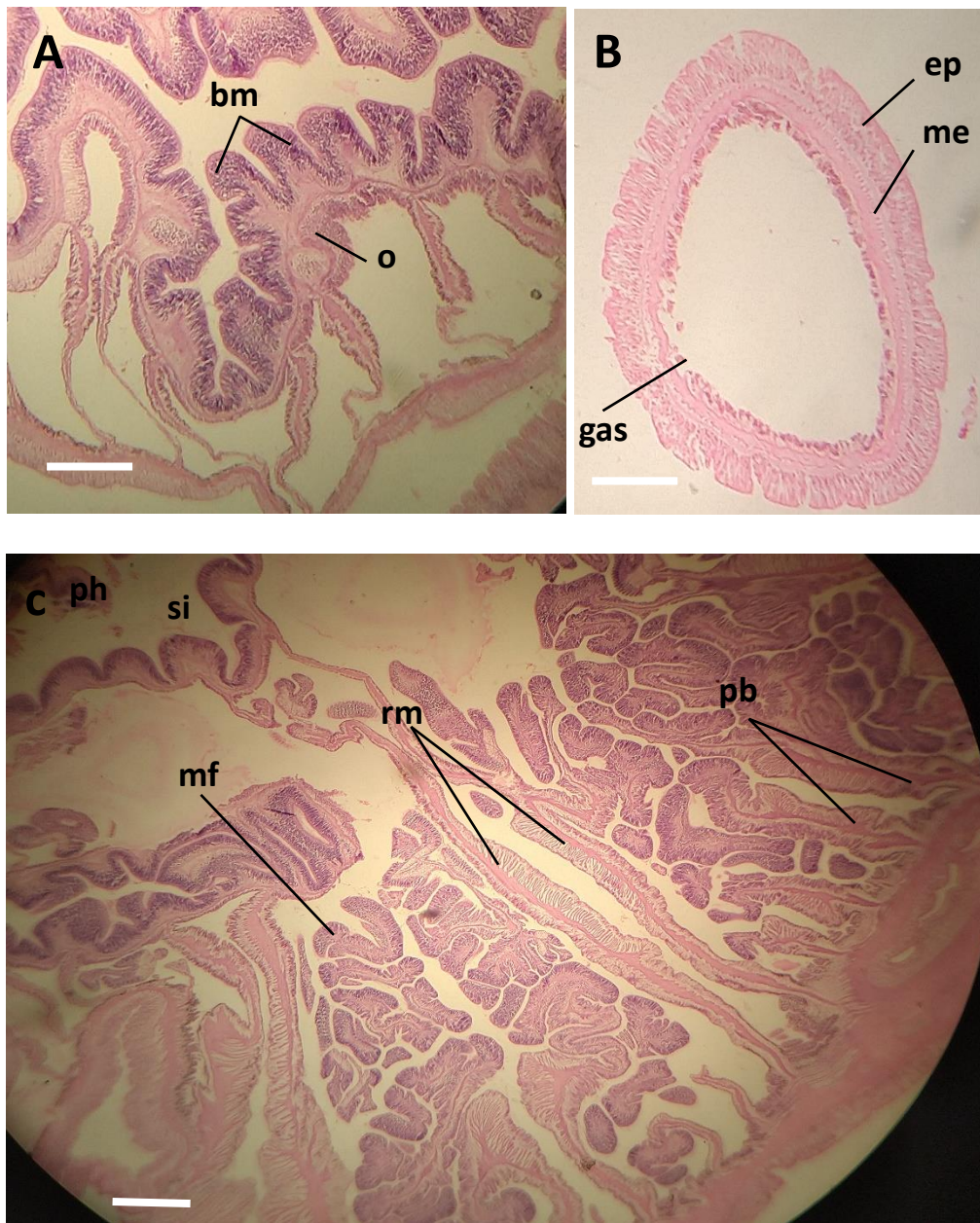


Plate 4.2: Histology of *Actinia equina* (Linnaeus, 1758).

A: Longitudinal section through base showing basilar muscles **B:** cross section through tentacles **C:** Transverse sections of proximal part of column. **bm:** basilar muscle **o:** ova **ep:** epidermis **me:** mesoglea **gas:** gastrodermis **ph:** actinopharynx **si:** siphonoglyph **mf:** mesenterial filaments **rm:** retractor muscle **pb:** basilar muscle parietobasilar muscle. Scale bar: 0.1mm

2. *Anemonia viridis* (Forsskal, 1775) (Plate: 4.3)

Synonyms:

Priapus viridis (Forsskal, 1775)

Actinia sulcata (Pennant, 1775)

Anthea cereus (Johnston, 18470)

Anemonia sulcata (Stephenson, 1935)

Common Name: Snake locks anemone

Material Examined: SHIAV01(22°28'41.80"N, 69°04'19.41"E) (CH-16 mm, CD-13 mm, LD-15 mm, PDD-22 mm, ODD-25 mm); SHIAV02 (22°28'42.51"N, 69°04'20.46"E) (CH-22 mm, CD-16 mm, LD-19 mm, PDD-23 mm, ODD-25 mm); SHIAV03 (22°20'10.96"N, 68°57'3.34"E) (CH-19 mm, CD-14 mm, LD-17 mm, PDD-21 mm, ODD-23 mm) collected from the intertidal zones of Shivrajpur (Kachhighadi), Dev Bhumi Dwarka District.

Description: This anemone inhabits the shallow sub littoral zone. Mainly found on the rocks where it exposed to the light and strong wave actions but also in sheltered places like bottom of the shallow intertidal pools. It can be growing up to 70 mm across the base with having tentacle span of 180-200 mm. The Column is smooth, with a row of discreet warts on the rim of the parapet. The colour of column is brownish or greyish. Species looks like the bunch of the tentacles arising from the substratum as the Column is often hidden under it. Base is slightly broader than the column. This anemone can be distinguished by long flowing and sinuous tentacles with green colour which found rarely retracted. Snake locks anemones hardly ever retract their tentacles due to the fact of the algae's want for sunlight. Even when left on land after high tide the tentacles remain exposed. The irregularly arranged tentacles can be up to 200 in numbers. Oral disc is white in colour with pattern of white radial lines. The *A. viridis* is oviparous (producing young by means of laid the eggs outside the mother's body. such sexual reproduction is less common than asexual longitudinal fission process (Horton, 2000 and Sick, 1991).

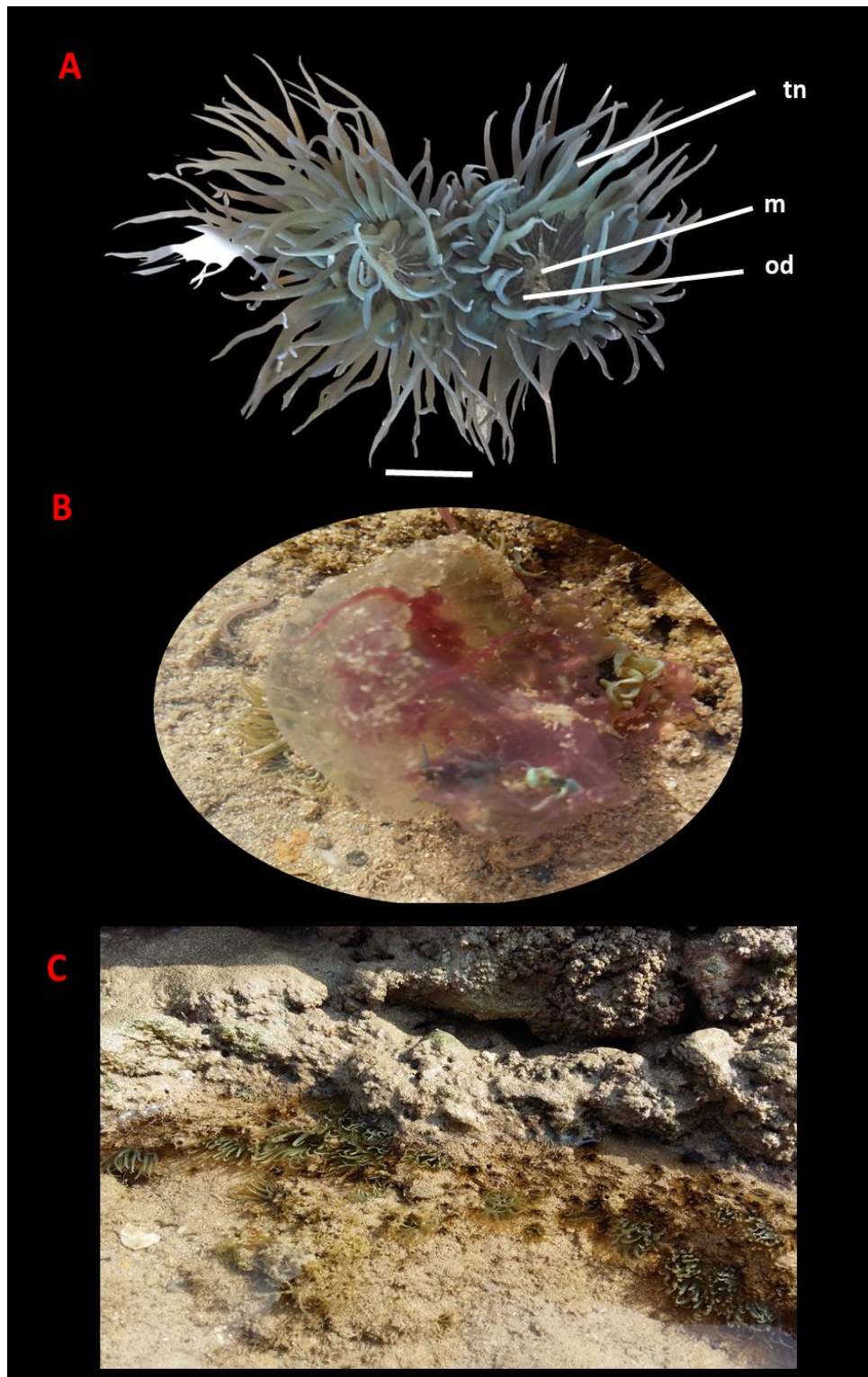


Plate 4.3: *Anemonia viridis* (Forsskal, 1775) A: Oral view of living specimen B: Specimen feeding behaviour C: specimen in living habitat. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

Distribution: The anemone is distributed through the Mediterranean Sea and north along Portugal, Spain and France to the southern and western coast of Great Britain. It is also found along the south of the straits of Gibraltar (African coast) (Horton 2000 and Sick, 1991). We reported this species from the Shivrajpur site facing Arabian Sea on west coast of Saurashtra.

3. *Anthopleura anjuna* Den (Hartog & Vennam, 1993) (Plate: 4.4)

Synonym:

Anthopleura midori (Uchida & Muramatsu, 1958)

Material Examined: VAJAA1 (20° 48' 52.488" N, 70° 31' 9.696" E) (CH-16 mm, CD-9 mm, LD-10 mm, PDD-12 mm, ODD-14 mm) Collected from the Vadodra jhala rocky shore.

Description: This anemone occurs from upper littoral to mid-littoral on rocky shores; is also present internally in shallow tide pools and wet crevices. Column of individuals can be darkish greenish brown with distinct, faded to whitish verrucae with a reddish central area. Column is entirely densely protected with conspicuous button- to cup-shaped verrucae, about 1-1.5 mm in cross-section, besides near the margin the place they are smaller. The verrucae are tremendously stalked and strongly adherent to overseas particles such as fragments of shells, massive sand grains, barnacles and calcareous worm tubes. In most specimens, their arrangement is obscured due to the state of contraction, but even in more or less extended specimens there are no distinct longitudinal rows, except distally, where short series of 3-6 smaller verrucae end marginally in a short more or less distinct marginal lobe, mainly with a distinct sub globular acrorhagus on its inner aspect. Oral disc having numerous creamy to yellow and some darkish radii. Directive radii white to cream, lining from the corners of the mouth to the directive tentacles and standing out against the darkish central area of the oral disc. Tentacles somewhat short, their number up to about 160 (120 in the examined specimen). Pattern of the tentacles can be diverse. Tentacles semi-translucent, creamy to greyish, above with opaque creamy spots and cross-bars. The absence of fission scars suggest

that the species does not reproduce asexually (Hartog and Vennam, 1993). More mesenteries are existing at the base than at the margin. Sphincter muscles are stronger, diffuse, hardly ever with a mild tendency to be meso-ectodermal (Plate 4.5).

Distribution: This species is described from west coast (Anjuna beach, Goa) of India (Hartog and Vennam, 1993). We have reported the distribution of this species from the rocky shore of Vadodra jhala site. This is the new record of sea anemone to the Gujarat coast.

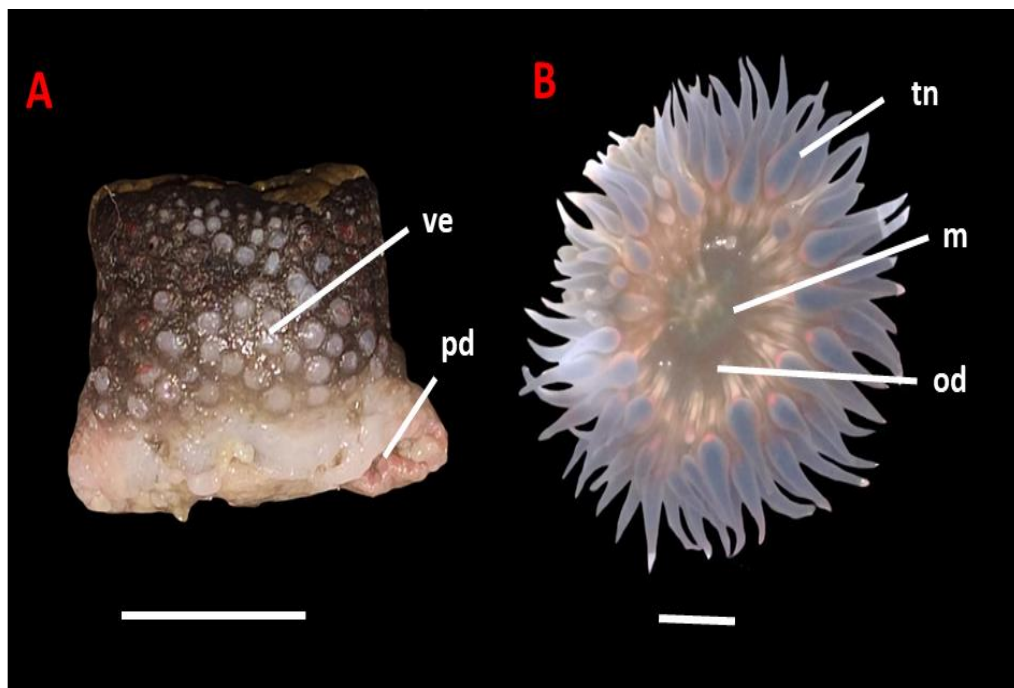


Plate 4.4: *Anthopleura anjunae* Den Hartog & Vennam, 1993.

A: Preserved specimen B: Oral view. Mouth (m), Oral disc (od), Verrucae (ve), Pedal disc (pd), Tentacles (tn). Scale bar: 10mm

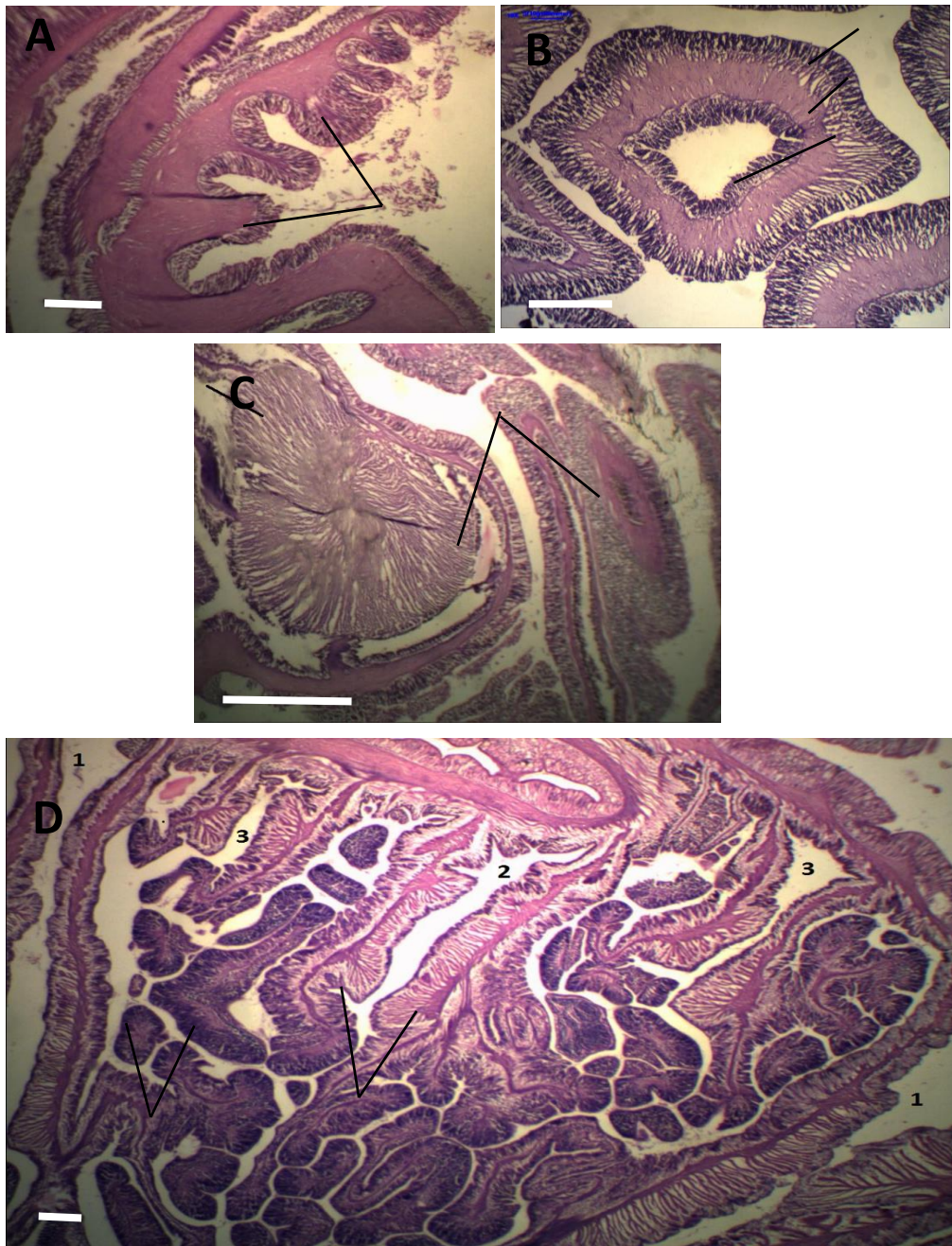


Plate 4.5: Histology of *Anthopleura anjuna* (Den Hartog & Vennam, 1993.)

A: Longitudinal section through base showing basilar muscles B: cross section through tentacles C: Section through marginal sphincter D: Transverse sections of proximal part of column. bm: basilar muscle ep: epidermis me: mesoglea gas: gastrodermis mf: mesenterial filaments rm: retractor muscle sp: sphincter muscle. Scale bar: 0.1mm

4. *Anthopleura artemisia* (Pickering in Dana, 1848) (Plate: 4.6)

Synonym:

Actinia Artemisia (Pickering in Dana, 1846)

Cereus Artemisia (Pickering in Dana, 1846)

Cibrina Artemisia (Pickering in Dana, 1846)

Common Name: Burrowing Anemone, Moon glow anemone

Material Examined: ALGAA01 (21° 22' 31.44" N, 72° 9' 57.888" E) (CH-50 mm, CD-40 mm, LD-45 mm, PDD- 47mm, ODD- 98mm) ALGAA02 (21° 22' 26.184" N, 72° 9' 53.964" E) (CH-35 mm, CD-30 mm, LD- 32mm, PDD- 35mm, ODD-78 mm) collected from the site of Alang, Bhavnagar.

Description: This anemone lives on open exposed coastlines often buried in mud or sand with only the crown exposed while the base is attached to solid substrate beneath the softer sediment. It can also have contracted into crevices and forms a low round-topped pillar. During the low tide, their presence is marked by a bump or puckered hole in the sand. Solitary polyps are around 25 mm in diameter and can be extended up to 70 mm. Fully expanded specimen can have crown and tentacle up to 40 mm in diameter and column expansion of 100 mm from its attachment to the sediment surface. The top third portion of the column is black or grey shading to white or pink at the bottom third. The polyp can have a very extended column with tubercles (verrucae) near the top, sparsely spaced and solitary on the middle of column and rarely found any verrucae on proximal 3rd of column (Fautin and Hand 2007). Verrucae are wart-like tubercle structures that pebble the column. They are adherent and collect a layer of shells and debris for protection. The column has a groove below the tentacles which is covered by a distinct fold or collar. Mesentery insertions as vertical white lines can be visible on the proximal third of column. The oral disc is broad, usually flat and can be red, brown, gray or black with solid or concentric patterns. It is about 1.5 times the column diameter when expanded. Its lips are not ribbed and do not extend beyond above the disc surface. The mouth is commonly an elongate slit. Tentacles can

be brown and green with presence of white colour band pattern. Tapering tentacles are about half as long as the oral disc diameter. There are irregular arrangements of tentacles having rarely more than 5 rows (Hand, 1955). The pedal disc is circular to irregular and is often wider than the column. There is no physa at the base. *Anthopleura artemisia* is the only species belong to this genus whose verrucae do not spread down to the base. This species has been observed in mass spawning events during low tide, in which every individual in the area releases their gametes in broadcast spawning (Weis *et al.*, 2002).

Distribution: The range is Alaska to southern California (Ricketts *et al.*, 1985) and possibly Japan (Hand, 1955). We reported this species from Alang.

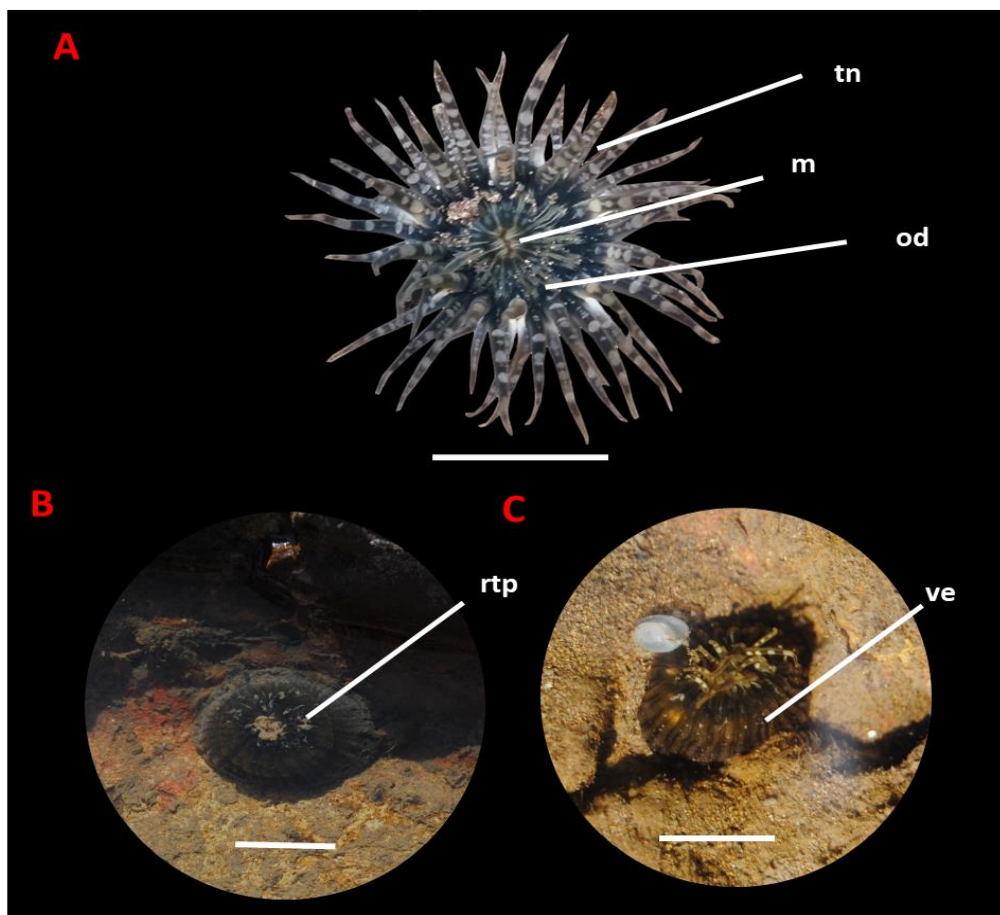


Plate 4.6: *Anthopleura artemisia* Pickering in Dana, 1848. A: Oral view of living specimen B, C: Contracted specimen. Mouth (m), Oral disc (od), Verrucae (ve), Tentacles (tn), rtp (Round top pillar). Scale bar: 10mm

5. *Anthopleura dixoniana* (Haddon & Shackleton, 1893) (Plate: 4.7)

Synonyms:

Actinioides dixoniana (Haddon & Shackleton, 1893)

Actinioides Papuensis (Haddon, 1898)

Actinogeton papuensis (Haddon, 1898)

Common Name: Banded bead Anemone

Material Examined: SUTAD01 (20° 49' 51.456" N, 70° 29' 13.164"E) (CH- 10mm, CD- 8mm, LD- 9mm, PDD- 10mm, ODD- mm) SUTAD02 (20° 49' 46.092" N, 70° 29' 15.576" E)(CH- 28mm, CD- 9mm, LD-11 mm, PDD- 14mm, ODD-15 mm) VAJAD03 (20° 48' 47.124" N, 70° 31' 14.448"E) (CH- 19mm, CD-13 mm, LD-15 mm, PDD- 17mm, ODD-29 mm),collected from Vadodra jhala and Sutrapada coast.

Description: This anemone crowded near the base of boulders and in crevices and cracks of the boulders at upper intertidal zone of rocky shore. These small anemones are often seen in clusters of many individuals packed close to one another, but not in aggregating pattern like many other *Anthopleura* spp. During the low tide, contracted anemones appear as shiny bumps on hard surfaces. Column, Oral disk and pedal disc diameter can be 6 mm and 8 mm long but usually expanded length about twice diameter. The entire anemone is usually in shades of brown and beige. The column is brownish or greyish near the base, greyish towards the margin with unpatterns grey verrucae. Verrucae are of same colour as column present only in distal part which may hold debris. The oral disk is raised around the mouth and relatively large compared to the tentacles. It is brown with green and yellow spots around the mouth and with cream and white patches scattered between mouth and base of tentacles which forms checkerboard pattern (Fautin, 2009). Tentacles are tapered and slender up to 60 arranged in 4-5 rows. Tentacles arranged in inner rows are longer than outer once and commonly solid white coloured on oral face. Outer tentacles are brownish with 2-4 white spots or bands along the length. Asexual

reproduction is commonly observed in *A. dixoniana*. It spawns once a year and divides asexually by longitudinal fission throughout the year (Lin *et al.*, 1992).

Distribution: Reported in Singapore from Pungol point, Pasir Panjang, Pasir Ris and Changi Creek (England, 1987). Distribution is also known from Australia, Hong Kong and the Maldives (Fautin, 2008). This is a new record to India through Sutrapada and Vadodra jhala of Gujarat Coast.

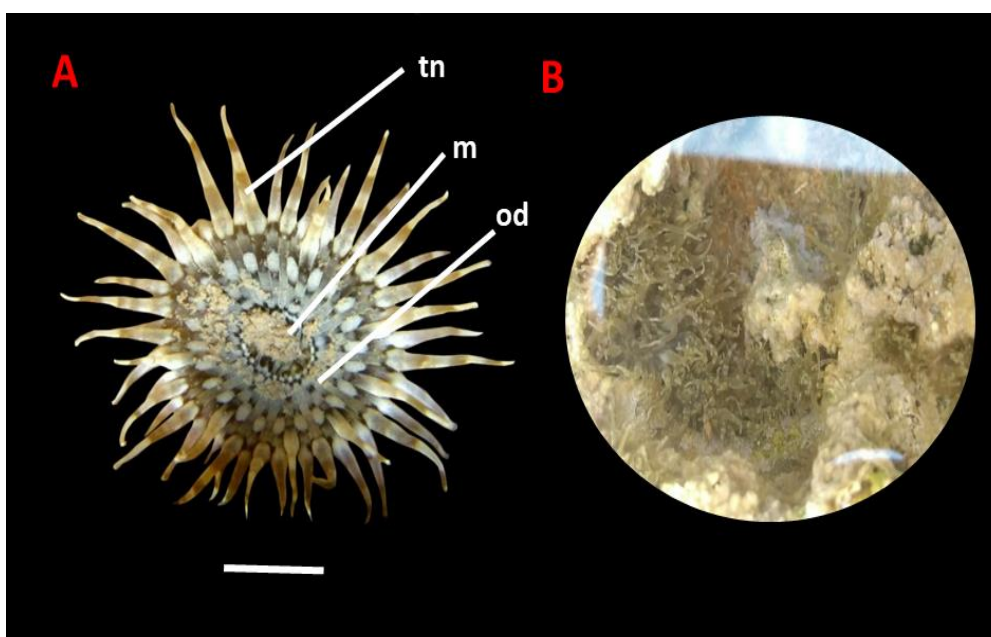


Plate 4.7: *Anthopleura dixoniana* (Haddon & Shackleton, 1893). A: Oral view of living specimen B: Specimen in natural habitat. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

6. *Anthopleura elegantissima* (Brandt, 1835) (Plate: 4.8)

Actinia (Taractostephanus) elegantissima (Brandt, 1835)

Bunoactis elegantissima (Brandt, 1835)

Cribrina elegantissima (Brandt, 1835)

Common Name: Green Aggregating anemone, Clonal anemone

Material Examined: VAJAE01 (20° 48' 51.948" N, 70° 31' 9.804" E) (CH-20mm, CD- 12mm, LD- 17mm, PDD-19 mm, ODD-20 mm), VAJAE02(20° 48' 50.76" N,

70° 31' 10.236" E) (CH- 14mm, CD- 9mm, LD-11 mm, PDD- 14mm, ODD- 13mm), VAJAE03(20° 48' 49.5" N, 70° 31' 10.92" E) (CH- 16mm, CD- 12mm, LD- 13 mm, PDD- 15mm, ODD- 17mm) ,VAJAE04(20° 48' 47.52" N,70° 31' 14.376" E) (CH- 11mm, CD- 7mm, LD-8 mm, PDD- 10mm, ODD-11 mm collected from Vadodra jhala coast.

Description: This anemone occupies exposed rocky habitats in mid littoral zone. It can be found either in dense populations or solitary. Such anemones often create large colonies consisting of closely packed hundreds of species. Algae mats formation in the intertidal creates hospitable, moist habitats for the aggregations (Niesen, 2007). Aggregating anemones can be 30mm long and wide up to 40mm. The column is usually pale grey- green, and sometimes shades to white at the base. When completely expanded, it is twice as long as wide. It becomes a hemispheric glob when contracted. The column is covered with vertical rows of adhesive tubercles (verrucae), create a layer of attached shells and debris. Verrucae are not densely packed and become fewer toward the base. The anemone has strong collar with well-developed fosse. The oral disc is broad and flat, with radiating lines (mesenterial insertions). It is somewhat wider than the column or of a similar width. The mouth is in the middle of the oral disc and the lips may be swollen with the surface of disc and are not ribbed. Tentacles are pointed having tipped with pink, lavender or white and about 1/4 as long as the diameter of the disc. There are more than 24 tentacles usually arranged in 5-6 rows (Fautin and Hand 2007). This species has a ring of white knobs, called acrorhagia present at just outside their ring of tentacles. The acrorhagia is filled with stinging cells and is used to kill other anemones. This anemone has a well-developed pedal disc that attached to the preferred substrate. The base is usually the same diameter as column. There are no bulbs at the base. There are both sexual and asexual reproductive cycles in this anemone. Anemone divides asexually via longitudinal binary fission, producing aggregations of "clones" common to this species which all are similar in colouration and sex (Hand, 1955; Fautin and Hand 2007). It may

possible that they settle higher in the intertidal and migrate lower to the tide-pools as they grow (Sebens, 1982b).

Distribution: Distribution range of this sea anemone is from Alaska to southern California (Hiebert, 2015). *Anthopleura elegantissima* is a new distribution record to India through Gujarat Coast.

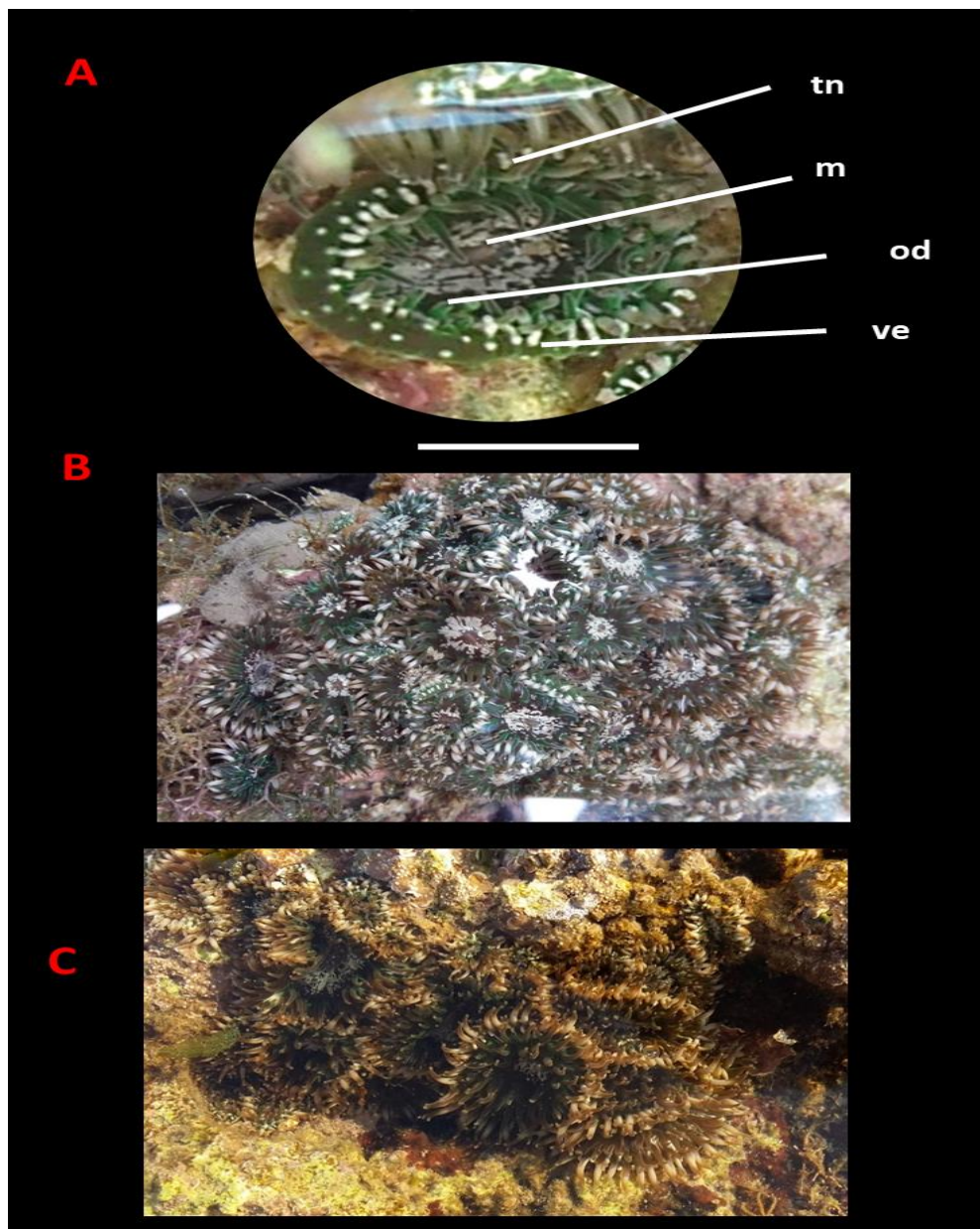


Plate 4.8: *Anthopleura elegantissima* (Brandt, 1835). A: Oral view of specimen B, C: colonial form of specimen. Mouth (m), Oral disc (od), Verrucae (ve), Tentacles (tn). Scale bar: 10mm

7. *Anthopleura sola* (Pearse & Francis, 2000) (Plate: 4.9)

Synonyms:

Actinia (Taractostephanus) elegantissima (Brandt, 1835)

Bunoactis elegantissima (Brandt, 1835)

Cribrina elegantissima (Brandt, 1835)

Cribrina elegantissima (Brandt, 1835)

Anthopleura elegantissima (Brandt, 1835)

Common Name: Solitary green anemone, Star bust anemone

Material Examined: SUTAS01 (20° 49' 36.9588" N, 70° 29' 20.76" E) (CH-20mm, CD- 12mm, LD- 14mm, PDD-16 mm, ODD-38 mm), SUTAS02 (20° 49' 40.9188" N, 70° 29' 18.7188" E) (CH- 23mm, CD- 13mm, LD-16 mm, PDD- 19mm, ODD- 40mm), VAJAS03(20° 48' 46.08" N, 70° 31' 15.564" E) (CH- 30mm, CD- 23mm, LD-26 mm, PDD- 28mm, ODD- 48mm), VAJAS04(20° 48' 51.192" N, 20° 48' 51.192" N)(20° 48' 50.976" N, 70° 31' 9.84" N) (CH- 22mm, CD- 12 mm, LD-15 mm, PDD- 18 mm, ODD-40 mm), VAJAS05 (20° 48' 52.74" N, 70° 31' 9.336" E) (CH- 23mm, CD- 13 mm, LD- 16 mm, PDD- 19 mm, ODD- 42 mm), VAJAS06 (20° 48' 52.776" N, 70° 31' 9.156" E)(CH- 19 mm, CD- 10 mm, LD- 13 mm, PDD- 16 mm, ODD- 36 mm) collected from the Sutrapada and Vadodra jhala Sites of Gujarat coast.

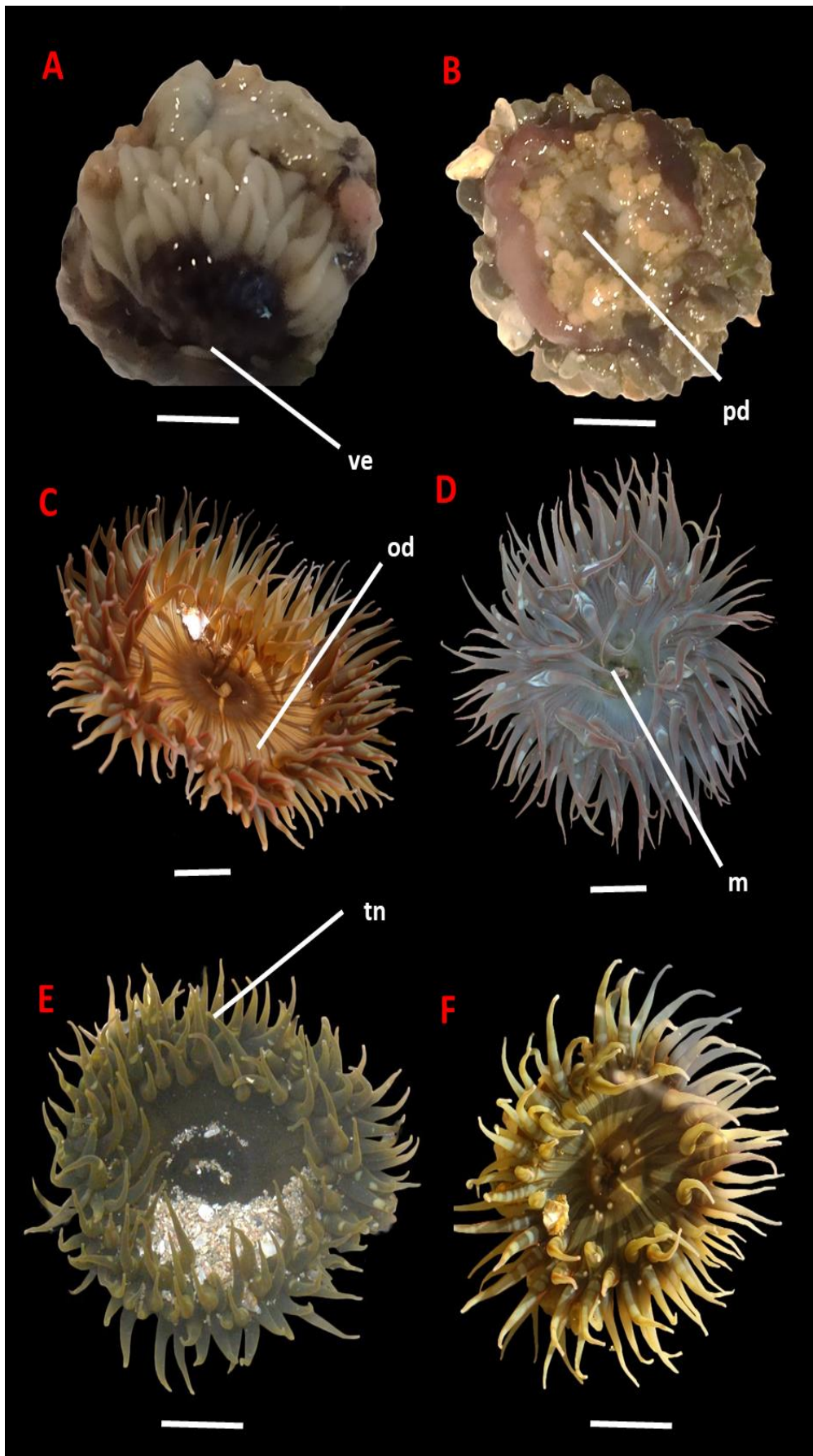
Description: This solitary sea anemone was previously considered as a non-clonal variant of the well-known, clonal Aggregating Anemone, *A. elegantissima*. Although polyps of these two species are similar in appearance and in cnidae, now recognised as a sibling species through documentation evidence of differences in population genetics, ecology, biogeography, development and life history (Pearse and Francis, 2000).

Anthopleura sola shares the morphological characters of *A. elegantissima*, except that *A. sola* is non-clonal and its polyps are large, growing to 200-250 mm across the tentacle crown. Large size is probably the critical factor which permits *A. sola* to inhabit subtidal depths that are beyond the vertical range of *A. elegantissima*. *A. sola* tends to thrive in somewhat sheltered positions and

moderate climates. On a microhabitat scale, they usually occupy protected places in tide pools, at the bases of boulders and in rock pockets and crevices. The oral disk is radially striped with wide variety of colour variations. The disc can be twice that size when extended. Pedal disc occupies a flat surface and can be relatively easily detached; a slender toe-like projection from the disc commonly retains a strong anchor-hold in a small hole in the rock, in which the young animal originally settled, long since outgrown. The column is green to white in colour and is twice as long as its width when extended. The column has numerous sticky protuberances arranged in vertical rows covered with gravels which serve as protection from desiccation and solar radiation and also provide camouflage against predators. The pointed and in colour olive to bright green short tentacles with pink or lavender tips arranged in five rings.

A. sola can reproduce both sexually and asexually, although most individuals reproduce asexually. *A. sola* are either male or female. The gonads are present in the gastro-vascular cavity, and eggs and sperm are released through the mouth. (Ricketts *et al.*, 1985 and Salinas, 2000). Because of its variability among species of genus *Anthopleura*, the morphology of a vesicle needs to be described in more detail than that of a verruca anatomically. Verrucae in particular this species is hollow evagination of the three layers of the column wall, with thick, glandular ectoderm that lacks nematocysts, relatively thin mesoglea (especially at the center, where it may form a cinclis), and relatively thin, un muscular endoderm (Plate: 4.10).

Distribution: *Anthopleura sola* is reported from the Pacific Ocean, along the west coast of North America from Alaska to Baja California. (Harbo, 1999). We have reported this species from Diu, Kodinar, Dhamlej, Veraval, Vadodra jhala and Sutrapada along the Southern Saurashtra coast of Gujarat.



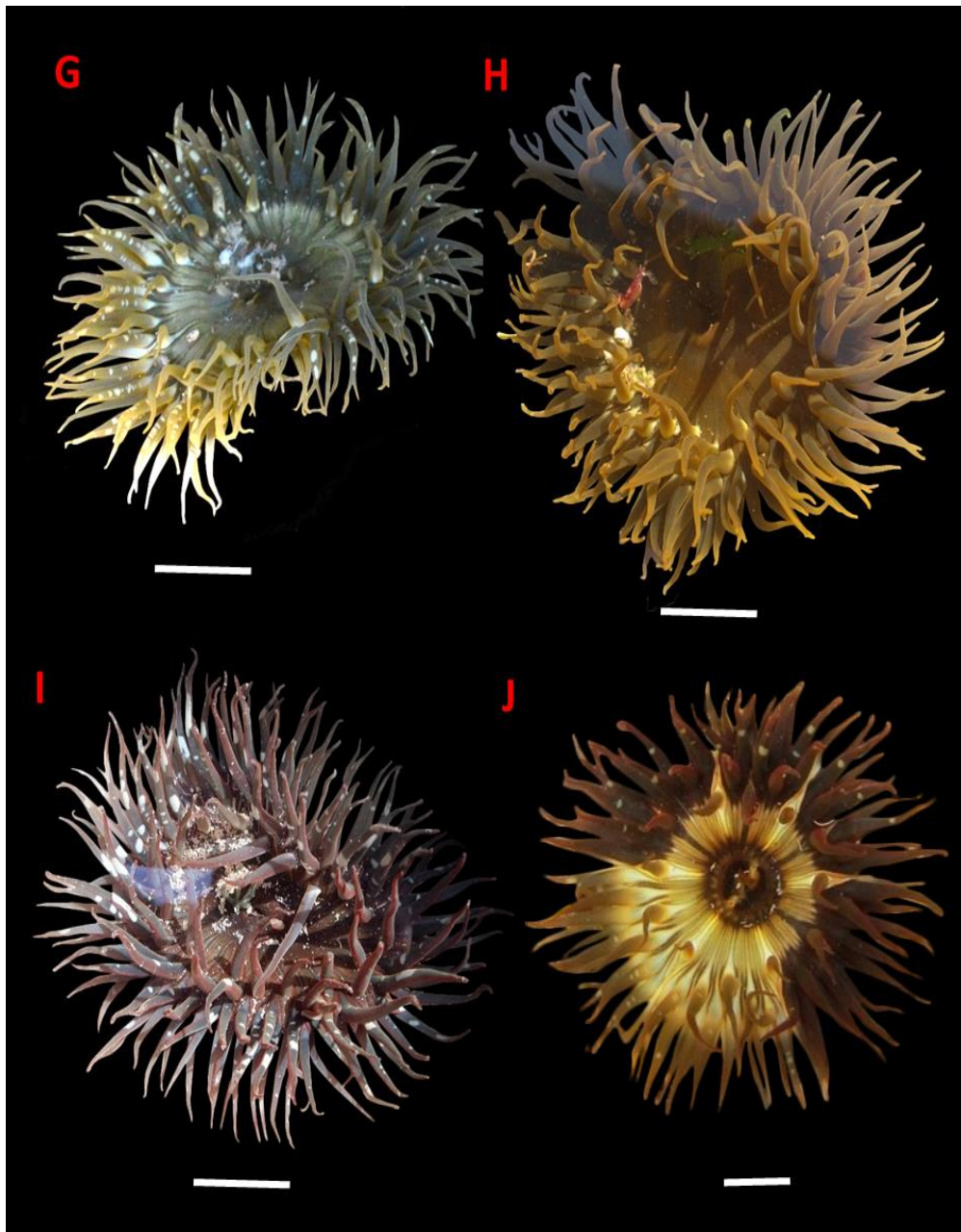


Plate 4.9: *Anthopleura sola* (Pearse & Francis, 2000). A: Preserved specimen B: Aboral view C-J: Oral view of morphs with variation in colour and tentacles. Mouth (m), Verrucae (ve), Pedal disc (pd), Oral disc (od), Tentacles (tn). Scale bar: 10mm

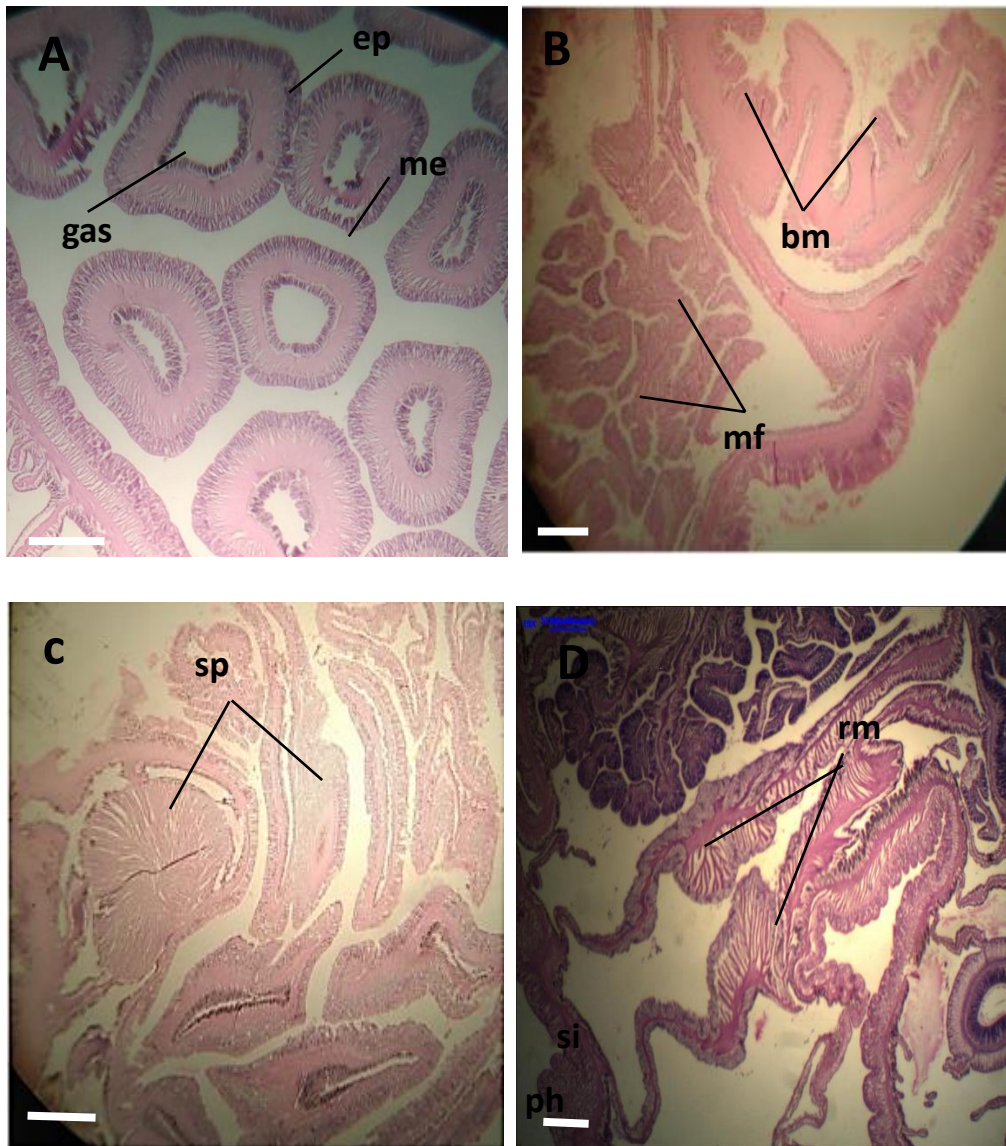


Figure 4.10: Histology of *Anthopleura sola* (Pearse & Francis, 2000).

A: cross section through tentacles B: Longitudinal section through base showing basilar muscles C: Section through marginal sphincter D: Transverse sections through column. bm: basilar muscle ep: epidermis me: mesoglea gas: gastrodermis mf: mesenterial filaments rm: retractor muscle sp: sphincter muscle. Scale bar: 0.1mm

8. *Macrodictyla doreensis* (Quoy & Gaimard, 1833) (Plate: 4.11)

Synonyms:

Actinia doreensis (de Blainville, 1830)
Actinia doreensis (Quoy & Gaimard, 1833)
Paractis doreensis (Milne Edwards, 1857)
Cereactis doreensis (Andres, 1833)
Condylactis gelam (Haddon & Shackleton, 1893)
Aulactinia gelam (Haddon, 1898)
Anthopleura gelam (Carlgren, 1949)
Radianthus gelam (Allen, 1972)
Macrodictyla gelam (Mariscal, 1972)
Radianthus gelam (Allen, 1973)
Radianthus malu (Moyer, 1976)
Macrodictyl adoreensis (Dunn, 1981)
Heteractis gelam (Cutress & Arneson, 1987)
Antheopsis gelam (Uchida & Soyama, 2001)

Common name: Long tentacle anemone and Corkscrew tentacle sea anemone

Material Examined: SUTMD01 (20° 50' 23.9994" N, 70° 29' 20.76" 70° 27' 43.2" E) (CH-37mm, CD-62mm, LD-65mm, PDD-70mm, ODD-95mm) collected from the Sutrapada coast.

Description: This anemone lives in mud, sand or gravel substrates with column partly buried in the substratum at Intertidal and sub tidal area. The base is circular and adheres to the substratum. The diameter of the base ranges from 50-100 mm. The base is the same colour as the lower column dull to brilliant red orange. Generally, the column is equal to or slightly larger than the basal diameter and smaller than the oral disk. The column's lower part is of uniform diameter and roughly equal to the pedal disk. In large specimens, the upper portion of the column is widely flared and only slightly extended in small specimens. The oral disc is widely flared. The oral disc is up to 500 mm diameter but is usually smaller and typically 100-200 mm diameter. Non-

adherent verrucae occur in longitudinal rows down the length of the column, are circular to eye-shaped and are the highest in the middle of the column with a diameter of up to 50 mm. The number of tentacles is relatively small but very long (up to 175 mm), sinuous and uniformly tapered to a point. The central intermediate portion of the column is usually creamy green and the top part of the column, the oral disk and the tentacles are grey-brown or brown-violet with white verrucae in longitudinal rows along the column walls. These rows can also extend as radial white lines to the oral disks and tentacles.

Distribution: Central Indo-Pacific in a narrow north-south band, from Japan south to the Philippines, Papua New Guinea, the Great Barrier Reef and Moreton Bay (Fautin *et al.*, 2008). From India, this species has been recorded from middle Andaman (Raghunathan *et al.*, 2014). We have reported this species from Veraval and Sutrapada coast.

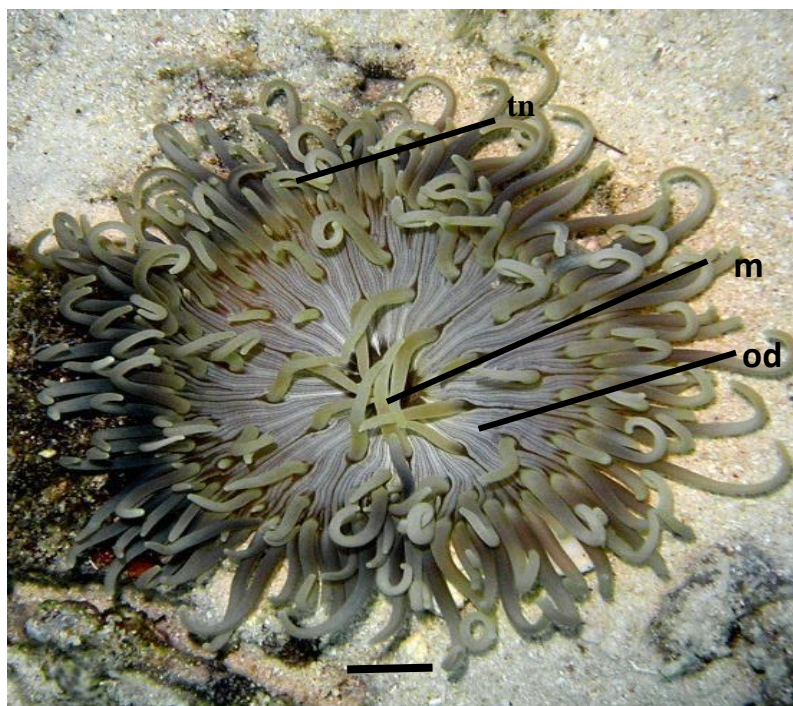


Plate 4.11: *Macrodactyla doreensis* (Quoy & Gaimard, 1833). Specimen present in the natural habitat. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

Genus *Urticina* (Ehrenberg, 1834)

Actiniidae having well-developed pedal disc. Column with adhesive or non-adhesive verrucae. Fosse well developed. Sphincter muscles strong, circumscribed. Tentacles short, stout, their longitudinal muscles ectodermal to mesogloal. Radial muscles of oral disc ectodermal to mesogloal. Numerous perfect mesenteries arranged as a rule decamerously or more irregularly 11, 12, 13 or 14 merously. Usually 10–20 oldest pairs sterile, rarely only six pairs. Same number of mesenteries proximally and distally (Sanamyan, 2006).

9. *Urticina clandenstina* (Sanamyan N., Sanamyan K. & McDaniel, 2013) (Plate: 4.12)

Common Name: Stubby rose anemone

Material Examined: SUTUC01 (20° 49' 40.9188" N, 70° 29' 18.24" E) (CH- 16mm, CD- 9mm, LD- 11mm, PDD-13 mm, ODD-16 mm), VAJUC02 (20° 48' 46.3212" N, 70° 31' 14.88" E) (CH- 20mm, CD- 12mm, LD-13 mm, PDD- 16mm, ODD- 19mm), VAJUC03(20° 48' 55.26" N, 70° 31' 4.1412" E) (CH- 13mm, CD- 7mm, LD-8 mm, PDD- 10mm, ODD- 13mm), VAJUC04(20° 48' 59.6412" N, 70° 31' 2.8812" E) (CH- 14mm, CD- 10 mm, LD-11 mm, PDD- 13 mm, ODD-15 mm), DWAUC05 (22° 14' 31.74" N, 68° 57' 20.7" E) (CH- 10mm, CD- 5 mm, LD- 7 mm, PDD- 9 mm, ODD- 11 mm), DWAUC06 (22° 14' 26.754" N, 68° 57' 26.6868" E)(CH- 11 mm, CD- 5 mm, LD- 6mm, PDD- 8 mm, ODD- 10 mm)

Description: This species is mostly found in the upper and middle intertidal zone, consisting of several tide pools that anatomize each other and form a large network of aquatic regimes as well as open rocks. It is always found buried with the pedal disk attached to solid objects such as buried stones or rock in coarse sand, empty gastropod shells and gravel, which thus shows both camouflage and as a deterrent to possible predators. Only the oral disk with the tentacles on the surface is visible. The column is cylindrical; typically the circular pedal adhesive disk is about the same as the column. The column has numerous adhesive verrucae that are present throughout its surface but in the distal half are more numerous and more prominent. The preserved specimens

hold a belt consisting of large and crowded gravel particles and broken shell firmly attached to column verrucae, although the region near the margin is nearly free of foreign matter. The pedal disc and column colour is usually red, sometimes pink, bright vermilion red in the holotype, darker in distal column; verrucae are of the same colour as the rest of the column without any markings of different colour. A deep fosse and short capitulum (a typical feature of many actiniids) are present. The oral disc is circular. The oral disc's background colour varies from bright to dull red, olive-green, gray, blue, or dirty yellow. The entire specimen photographed in situ have about 100 to 160 tentacles are visible. The tentacles are always arranged on the outer half of the oral disk decamerously in five cycles. They are short, sometimes extended, as long as half the diameter of the oral disk, but usually much shorter and thick, with tips of about the same length, the inner part may be slightly thicker than the outer part. Tentacle bases are outlined by short, thin red radial lines running on the oral disc. These lines may be quite well developed in some specimens feebly marked or even absent in others. The tentacles' background colour is similar to the oral disc's colour. Every tentacle at the base and in the middle has a transverse band coloured whitish or rose, encircling it completely or embracing only from the oral and lateral sides; these bands are divided by a broader reddish band. White bands can be V-shaped on the oral sides of the tentacles. The sexes are separate; there were no embryos in the specimen being examined. The sphincter muscles generally have one center lamella which varies considerably in shape and degree of the development (Plate: 4.12). The endoderm thickness of the siphonoglyphs is 5 times thicker than other parts of actino-pharynx while the thickness of mesoglea is about same as rest of the parts.

Distribution: its range extents from south Alaska to California (Sanamyan, 2013). We have reported this species from the southern and eastern end of Saurashtra coast of Gujarat.

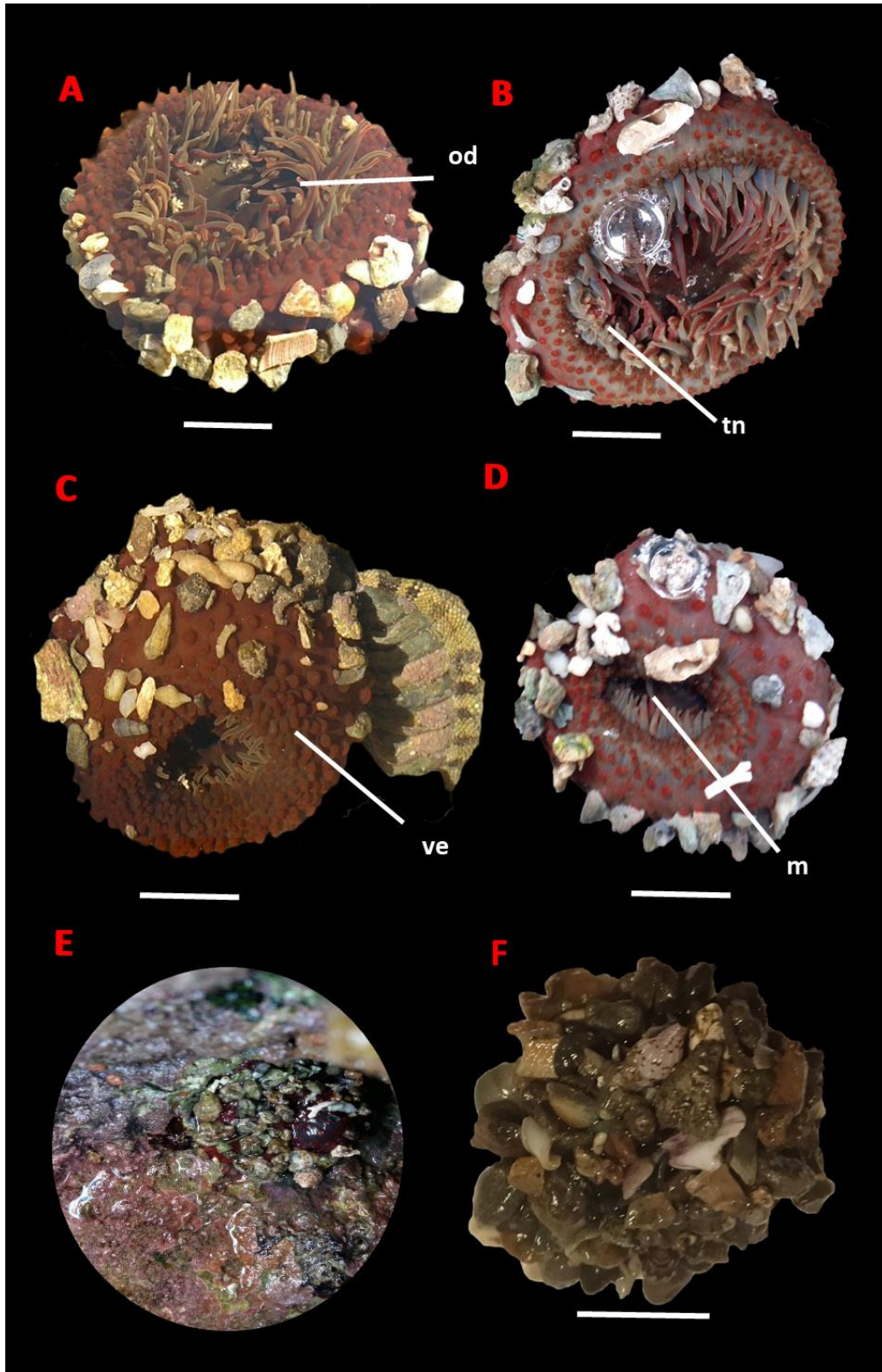


Plate 4.12: *Urticina clandenstina* (Sanamyan N., Sanamyan K. & McDaniel, 2013). A, B, C, D: Oral view and contracted specimen E,F: Specimen showing camouflage mechanism. Mouth (m), Oral disc (od), Verrucae (ve), Tentacles (tn). Scale bar: 10mm

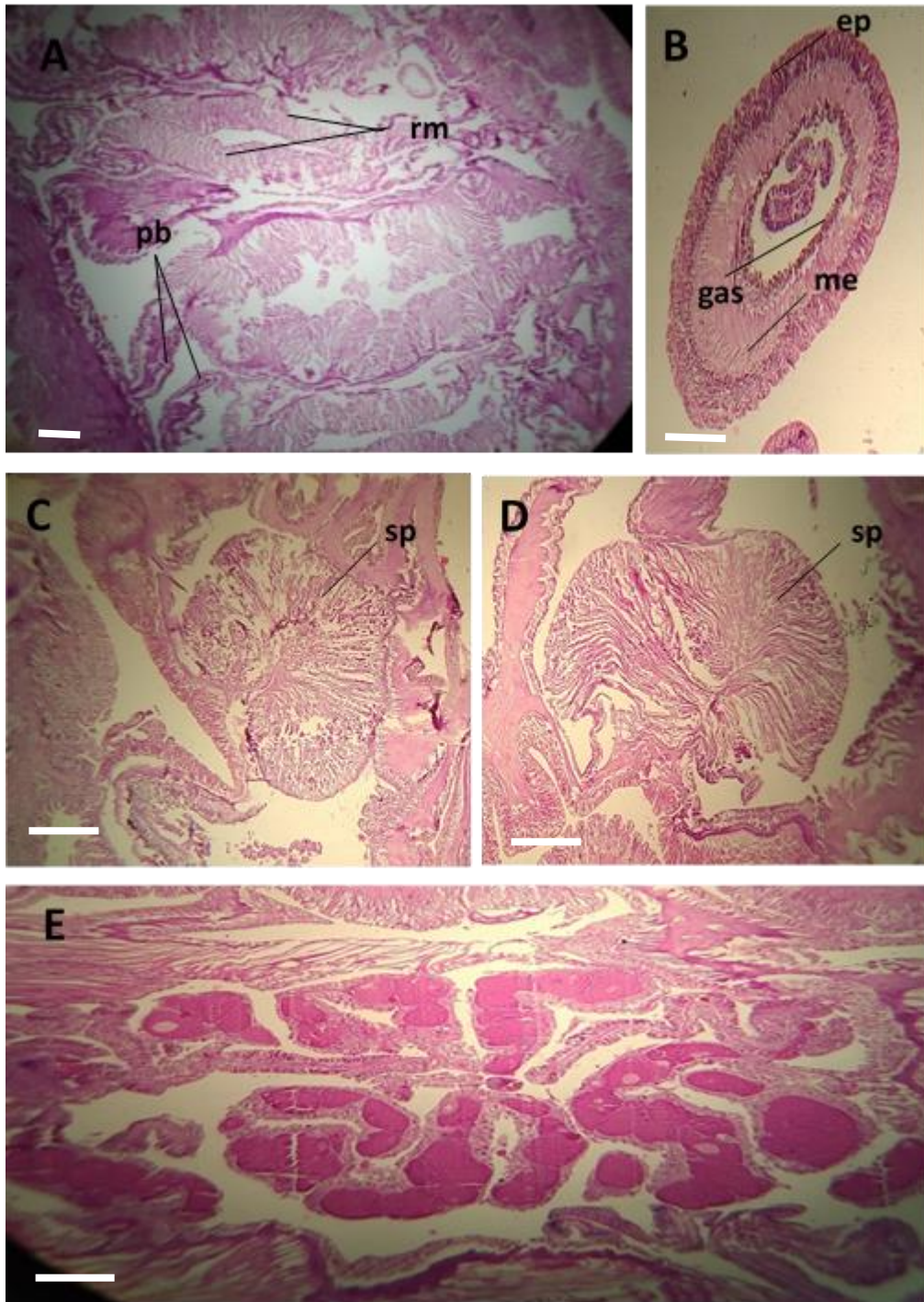


Plate 4.13: Histology of *Urticina clandenstina* (Sanamyan N., Sanamyan K. & McDaniel, 2013). A: Transverse sections of proximal part of column B: cross section through tentacles C, D: Section through marginal sphincter E: Gametogenic region of mesentery of female specimen. ep: epidermis me: mesoglea gas: gastrodermis rm: retractor muscle pb: parietobasilar muscle sp: sphincter muscle. Scale bar: 0.1mm

Family Aiptasiidae (Carlgren, 1924)

Aiptasiidae with elongate, smooth column. Cinclides arranged in girdle around centre of body. Sphincter mesogleal, very weak. Margin tentaculate. Tentacles long, simple, and without projections. Siphonoglyphs present. Six pairs of perfect mesenteries. Gonads on mesenteries of first cycle and on strongest imperfect ones. Acontia well developed (Carlgren, 1949).

Genus *Exaiptasia* (Grajales & Rodriguez, 2014)

Aiptasiidae with pedal disk adherent. Column elongated, flat, with cinclides in mid-column in 2–3 longitudinal rows; column not clearly divided into scapus and capitulum. Mesogleal marginal sphincter moderately strong. Strong ectodermal longitudinal muscles in column distal. Long, simple, up to 96 tentacles, always smooth, with no projections. The same number of distal and proximal mesenteries. Six perfect pairs of mesenteries. First and second fertile cycles. Restricted muscles of the retractor. Well-developed Acontia. (Grajales, 2014).

10. *Exaiptasia diaphana* (Rapp, 1829) (Plate: 4.14)

Synonyms:

Aiptasia diaphana (Rapp, 1829)

Aiptasia inula (Duchassaing & Michelotti, 1864)

Aiptasia leiodactyla (Pax, 1910)

Aiptasia mimosa (Duchassaing & Michelotti, 1864)

Aiptasia minuta (Verrill, 1867)

Aiptasia pallida (Agassiz in Verrill, 1864) (new genus created by Grajales & Rodriguez 2014)

Aiptasia pulchella (Carlgren, 1943)

Aiptasia saxicola (Andres, 1881)

Aiptasia tagetes (Duchassaing & Michelotti, 1864)

Aiptasiomorpha (*diaphana* Rapp, 1829)

Aiptasiomorpha leiodactyla (Pax, 1910)

Aiptasiomorpha minuta (Verrill, 1867)

Exaiptasia pallida (Agassiz in Verrill, 1864 (subjective synonym; epithet diaphana retains priority over pallida (ICZN Opinion 2404))

Paranthea minuta (Verrill, 1867)

Common name: Glass Rose Anemone, Trumpet Anemone

Material Examined-VAJAD01 (20° 49' 48.8382" N, 70° 29' 16.5006"E) (CH-10mm, CD- 8mm, LD - 9mm, PDD-10 mm, ODD-12 mm), SHIAD02 (22° 14' 34.76" N, 68° 57' 17.65" E) (CH-20mm, CD- 14mm, LD - 16mm, PDD-17 mm, ODD-30 mm), SHIAD03 (22° 14' 32.34" N, 68° 57' 22.71" E) (CH-30mm, CD- 9mm, LD - 11mm, PDD-14 mm, ODD-11 mm) collected from the Vadodra jhala and Shivrajpur coast.

Discription: *Aiptasia* anemones are found along protected coasts and intertidal rocky shorelines in shallow waters. They find themselves attached to rubble alone, live rock, dead corals, and other hard substrates. They also form dense colonies in shallow water areas, sometimes as dense as solid sheets. Oral disc can be upto 40mm in expanded specimen. *Aiptasia diaphana* is somewhat transparent and typically appears in a brownish or pink to reddish colour with lighter tips. Tentacles up to 96, smooth, long to tips tapering, all of the same length, up to 20 mm. clear filament appearance with curved acute point traverses them. Its peristome is circular and wider than the column with large mouth opening. A pedal disk with a diameter of 10 mm, larger than the base. Base smooth to 60 mm in preserved specimens and to 30 mm in diameter. This species shows both sexual and asexual reproduction through pedal laceration (Hunter, 1986), giving us a model that can be easily manipulated in the laboratory. Retractor muscles are restricted. Sphincter, mesogleal are very weak (Plate: 4.15).

Distribution: Its distribution is mainly along the Mediterranean Sea, Atlantic Channel and North Sea. It also occurs from Portugal to the Canary Islands and has been introduced into the Red Sea, Gulf of Aqaba (Gosliner *et al.*, 1996). We have reported this species throughout the Saurashtra coast.

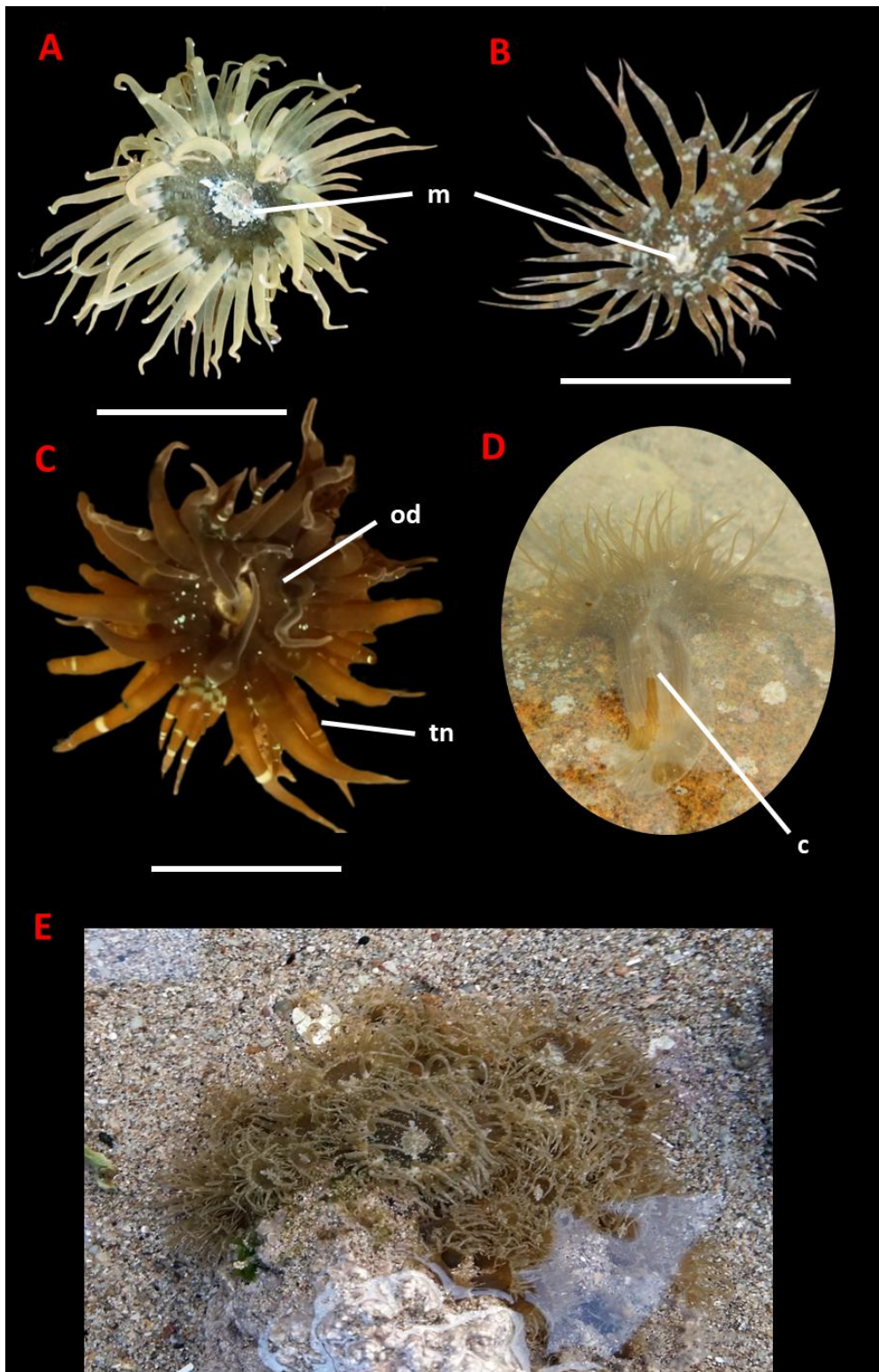


Plate 4.14: *Exaiptasia diaphana* (Rapp, 1829). A, B, C: Oral view of specimen D: Expanded column E: specimen in natural habitat. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

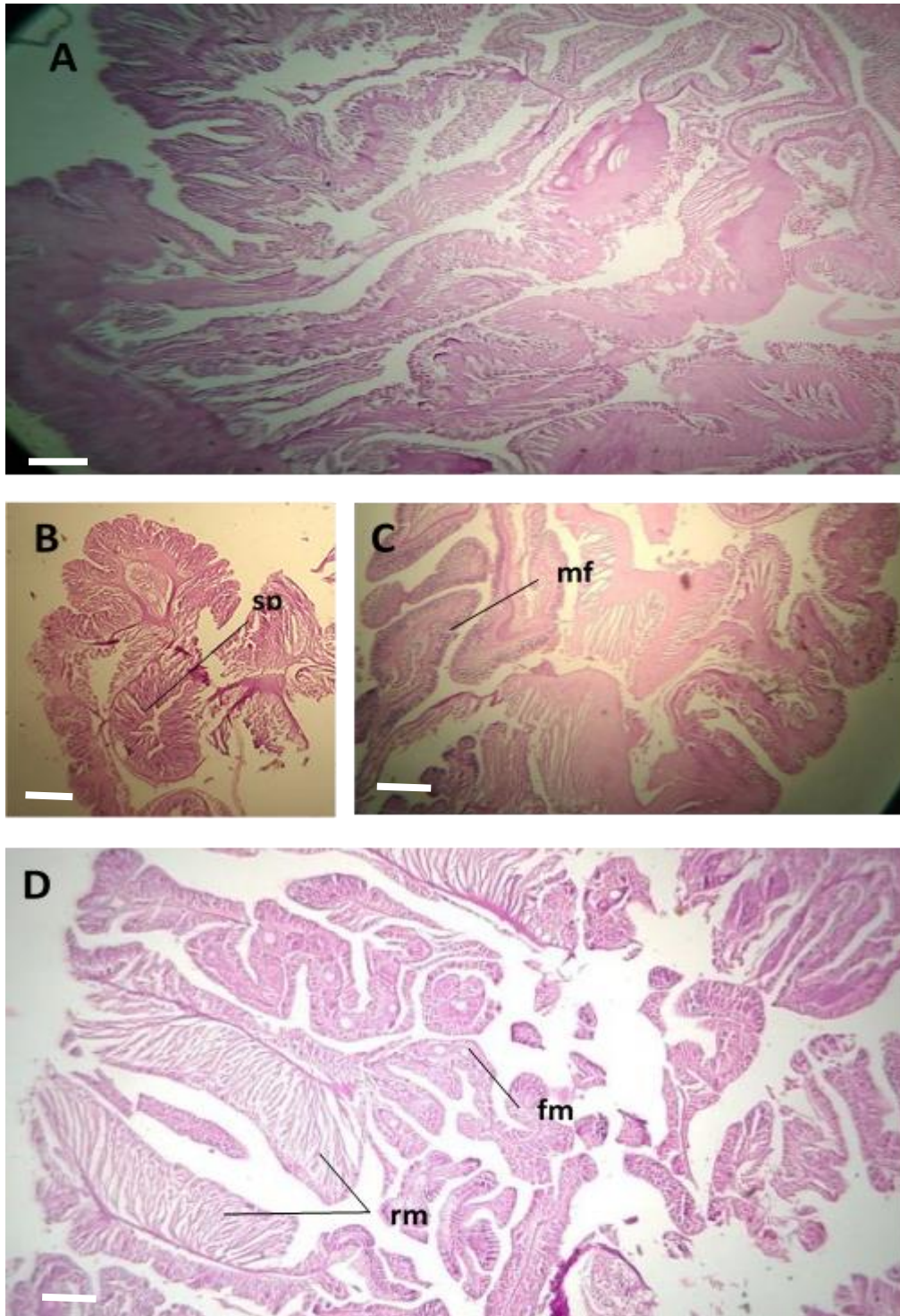


Plate 4.15: Histology of *Exaiptasia diaphana* (Rapp, 1829).

A: Transverse sections of proximal part of column showing mesenterial arrangements **B:** Section through marginal sphincter **C, D:** Transverse sections through column. **mf:** mesenterial filaments **rm:** retractor muscle **sp:** sphincter muscle **fm:** female fertile mesentry. Scale bar: 0.1mm

Family Diadumenidae (Stephenson, 1920)

Present Acontia, No distinct sphincter. Some or all of the inner tentacles that usually form catch-tentacles supplied with atrichs and holotrichs; often, it seems, there is a lack of these unique nematocysts in individuals of a species that possesses them (Cutress, 1949)

Genus *Diadumene* (Stephenson, 1920)

Well-developed Acontia. Smooth base, which can be divided into scapus and capitulum. Well-developed oral disc. The margin of the tentaculate capitulum. No separate number of sphincter. Tentacles; imperfectly retractile. Some or all of the inner tentacles are thicker, forming atrich and holotrich catch-tentacles. Variable number of siphonoglyphs and directives. Perfect mesenteries, six or more pairs; Perfect mesenteries, six or more pairs; fertile ones perfect and stronger. The circumscript-diffuse retractor. Weak muscles of the parietobasilar and basilar (Cutress, 1949).

11. *Diadumene lineata* (Verrill, 1869) (Plate: 4.16)

Synonyms:

Aiptasiomorpha (*Diadumene*) *luciae* (Verrill, 1899)

Diadumene luciae (Verrill, 1869)

Haliplanella lineata (Verrill, 1869)

Haliplanella luciae (Verrill, 1898)

Sagartia lineata (Verrill, 1869)

Sagartia luciae (Verrill, 1898)

Common name: Striped Sea Anemone, Orange Striped Green Anemone

Material Examined: SUTDL01 (20° 48' 54" N, 70° 30' 54" E) (CH-20mm, CD-18mm, LD-19mm, PDD-21mm, ODD-27mm) collected from Sutrapada coast.

Description: *Diadumene lineata* inhabits the micro pools and tide pools of intertidal and shallow subtidal areas, both on solid substrates and on roots and stems in marshland protected areas. In literature, it is frequently found in places near harbours, pilings, and floats or shipping lanes. Also occurs in

brackish-water creeks and lagoons, in estuaries, etc. (Molina *et al.*, 2008). It is a small anemone, usually less than 40 mm in height, with an approximately the same size tentacular crown. The column of the body is smooth and cylindrical. The most common, distinctive and attentive colour morph has a greenish brown column with orange stripes (there are 12 stripes in regular shapes, but the number of stripes may be irregular (Haussermann *et al.*, 2015); other varieties have 48 paired white stripes or in rare cases the stripes are absent (Shick & Lamb, 1977). The tentacles are tapering, transparent with a white coloured base. Usually there are 50-60 (and as many as 100) tentacles. Within their native range. *D. lineata* reproduced sexually while introduced Individuals reproduce mostly through longitudinal fission and sometimes through pedal laceration (Shick and Lamb, 1977; Molina *et al.*, 2009).

Distribution: *Diadumene lineata* was originally described in Japan but was recorded as an invasive species from the world's temperate and tropical coasts, mostly transported by ships or seafood shipments. It has distributed and reported Pakistan, China, Hong Kong, Indonesia, China, Japan, Ireland, North America, Canada, Hawaii, New Zealand (DeFelice *et al.*, 2001; Cairns *et al.*, 2003; Minchin, 2007 and Kazmi,2016). We have documented this species from Veraval and Saurashtra coast.

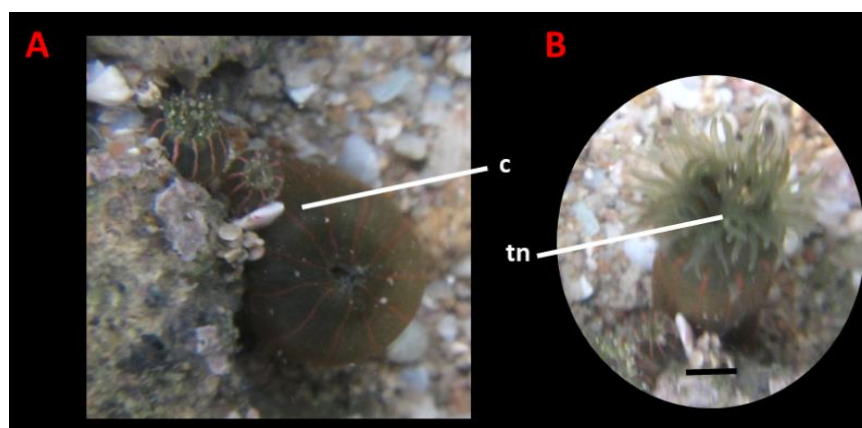


Plate 4.16: *Diadumene lineata* (Verrill, 1869). A, B: Specimen in natural habitat. Column(c), Tentacles (tn). Scale bar: 10mm

Family: Phymanthidae (Andres, 1883)

Soft column, Suckers on lower portion increasing in size from below upwards, Found in clusters attached to beneath branching corals in shallow water; crenulated parapet. mouth rounded, with two grooves; two types of tentacles: centripetal and peripheral; centripetal tentacles short, conical, arranged in three cycles; peripheral tentacles arranged in four or five cycles.

Genus *Phymanthus* (Milne Edwards & Haime, 1851)

Phymanthidae with base and column of variable appearance. Upper part of column with verrucae, which sometimes may be indistinct. A row of perforated marginal pseudospherules Cinclides may be present at the base. No sphincter or a very weak, diffuse one. Marginal tentacles arranged hexa- or octomerously. Laterally the marginal tentacles are provided with weak or well-developed protuberances in the form either of low knobs or of ramified branches. Longitudinal muscles of tentacles and radial muscles of oral disc ectodermal, sometimes with a slight tendency to be meso-ectodermal. Pairs of mesenteries arranged hexa- or octomerously, many are perfect, the stronger with or without the directives, fertile. Retractors well developed, strong, diffuse or strongly restricted, reniform.

12. *Phymanthus crucifer* (Le Sueur, 1817) (Plate: 4.17)**Synonyms:**

Cereus crucifer (Le Sueur, 1817)

Actinia crucifera 9Le Sueur, 1817)

Phymanthus cruciferus (Andres, 1883)

Ragactis cruciata (Andres, 1883)

Phymanthus crucifer (McMurrich, 1889)

Epicystis crucifera (Verrill, 1898)

Epicystis osculifera 9Verrill, 1900)

Phymantes crucifer (Cutress & Cutress, 1976)

Phimanthus crucifer 9Zamponi, 1981)

Epicystis crucifer (Cairns, den Hartog, & Arneson, 1986)

Common name: Rock flower anemone, red beaded anemones

Material Examined: OKHPC01 (22° 28' 51.06" N, 69° 5' 15.108" E) (CH-31mm, CD-40mm, LD-41mm, PDD-43mm, ODD-63mm) collected from Okha coast of Gujarat.

Description: They inhabit the sandy bottoms of the sea and is found approximately 1-5 m deep in the ocean. The main part (column) of the anemone is usually buried in the sand, anchored to a rock below the surface, allowing the anemone to pull back into the substrate when disturbed. Documented as a species frequently associated with coral reefs and rocky ledges, when agitated, *P. crucifer* can withdraw into crevices and holes. Fully expanded specimen with a diameter of 40–100 mm oral disc. Oral disks can usually be up to 32–55 mm in diameter, rough, colour variable, often white with dark brown, white with olive-green or grey with green and brown. Marginal and distal tentacles: short marginal tentacles, about 350–360, distally tapering, with annular or smooth thickenings, olive or light brown with longitudinal coloured stripes. Cylindrical column is smooth, diameter 27–45 mm and height 15–35 mm. The base of the column is generally cream-coloured with stripes of red, graying upward. Rows of light and dark stripes and bumps radiate outward from the mouth, varying in colour from bright green in the center to brown, lavender, yellow or white outward. It has bright red suckers on its column, to which debris can be attached for camouflage purposes. Generally developed pedal disk is with a diameter of 12–35 mm, bright pink or orange. The red beaded anemone is a dioeciously species, meaning separate individuals are male and female.

Distribution: *Phymanthus crucifer* is found along the Caribbean Sea, from Bermuda to Barbados (Gonzalez-Munoz et al., 2012). We have reported this species from Okha coast.

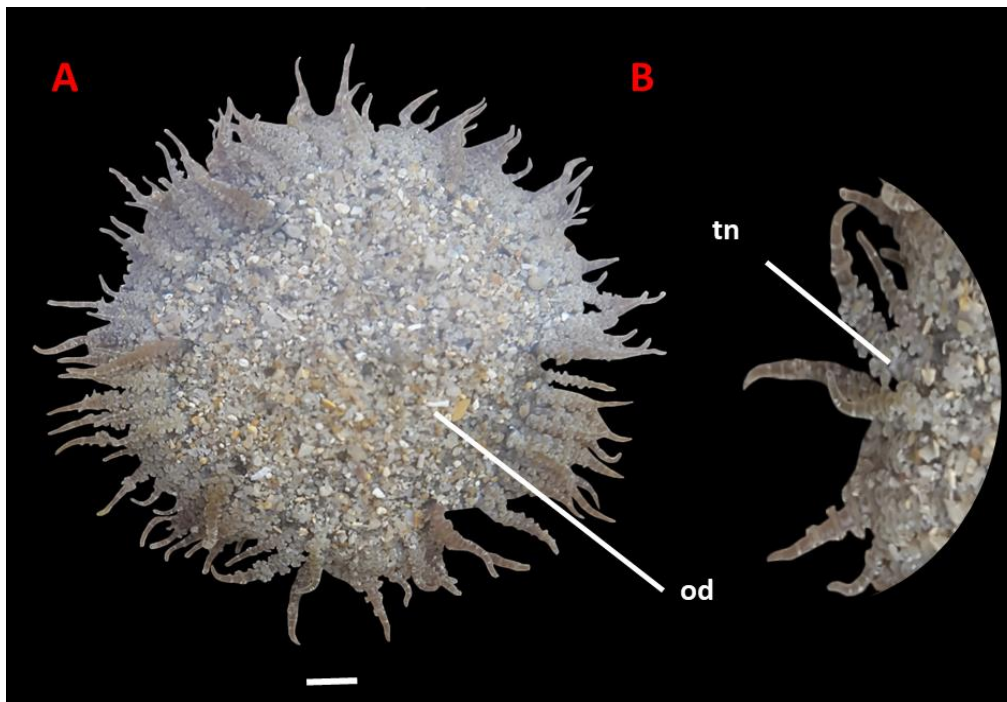


Plate 4.17: *Phymanthus crucifer* (Le Sueur, 1817). A: Oral view of specimen B: Tactile details. Oral disc (od), Tentacles (tn). Scale bar: 10mm

13. *Phymanthus buitendijki* (Pax, 1924) (Plate: 4.18)

Material Examined: OKHPB01 (22° 28' 51.06" N, 69° 5' 15.108"E) (CH-30mm, CD-22mm, LD-26mm, PDD-32mm, ODD-70mm) collected from Okha.

Description: This species inhabits the sandy bottoms. The column is flat, inconspicuous, and the colour is bluish grey. The oral disc is dark greenish blue, the mouth is in most species on the tip, and the oral disc is lined with sediments. Tentacles are arranged at the periphery of the oral disc in two or three orders. Tentacles are long, tapered tips, small flower like projections attached directly to the tentacles, alternatively arranged, highly branched and brightly coloured. Purple lines between the end of the oral disc and the tentacle. Tentacles are mostly curved inside that looks like a fishing hook.

Distribution: This species recorded distributed from Indonesia, Andaman Nicobar island (Raghunathan *et al.*, 2014). We have documented this species from Okha coast.

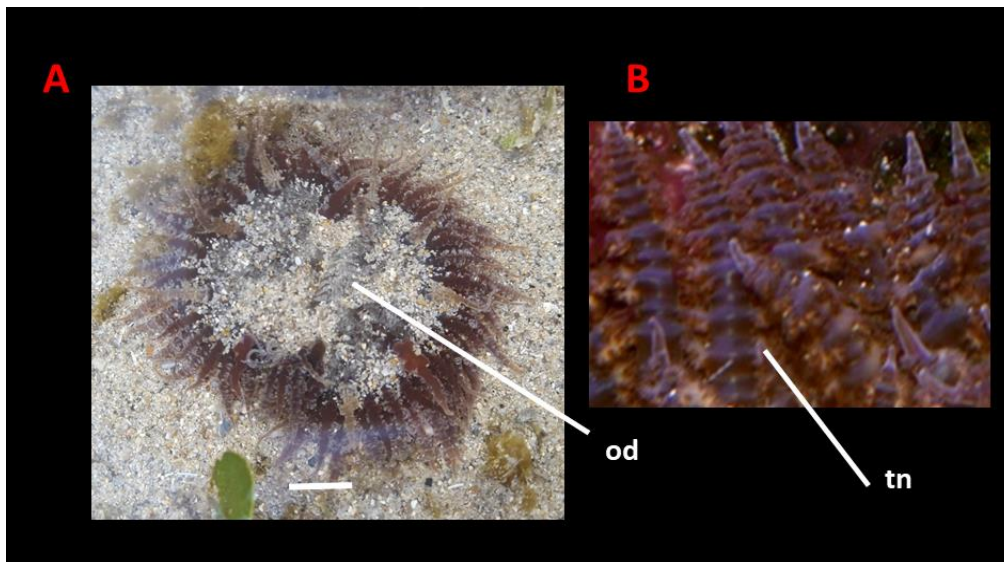


Plate 4.18: *Phymanthus buitendijki* (Pax, 1924) A: Specimen buried in sandy habitat B: Tentacle details. Oral disc (od), Tentacles (tn). Scale bar: 10mm

Family Stichodactylidae (Andres, 1883)

Column usually with verrucae. Endomyaria with well-developed pedal disc. Tentacles are short and wart like, never very long. From all or some endocoel tentacles arise in radial series, sometimes more than one row per endocoel or only one, rarely radial arrangement indistinct or absent. Only one tentacle communicates with each exocoel. Sphincter muscles diffuse to circumscribed, usually not very strong. Retractor muscles weaker or stronger, diffuse. Longitudinal muscles of tentacles and radial muscles of oral disc ectodermal (Song and cha, 2004).

Genus *Heteractis* (Milne Edwards, 1857)

Well-developed pedal disc, adherent, slightly wider than the lower column, narrower than the oral disk. Smooth proximal column, distally with to prominent verrucae unnoticeable. Fosse is absent in the depths. Difficult to retractile oral disk, smooth to shallow undulating. Tentacles are all alike, sinuous to digitiform on one individual; one specie has swellings on lateral and

oral surfaces. More than one tentacle per endocoel is usually considered. (Fautin *et al*, 2008).

14. *Heteractis crispa* (Ehrenberg, 1834) (Plate: 4.19)

Actinia (Entacmaea) crispa (Hemprich & Ehrenberg in Ehrenberg, 1834)

Actinia paumotensis (Couthouy in Dana, 1846)

Discosoma macrodactylum (Haddon & Shackleton, 1893)

Discosoma tuberculata (Kwietniewski, 1898)

Radianthus kukenthali (Kwietniewski, 1896)

Radianthus lobatus (Kwietniewski, 1898)

Stoichactis tuberculata (Stephenson, 1922)

Common name: Leathery sea anemone, Sebae anemone

Material Examined: OKHHC01 (22° 28' 52.4994" N, 69° 4' 42.78" E) (CH-49mm, CD-30mm, LD-35mm, PDD-45mm, ODD-90mm), OKHHC 02 (22° 28' 50.2788" N, 69° 4' 46.0806" E) (CH-29mm, CD-23mm, LD-25mm, PDD-30mm, ODD-38mm) OKHHC03 (22° 28' 49.1262" N, 69° 4' 48.036" E) (CH-40mm, CD-36mm, LD-40mm, PDD-46mm, ODD-94mm) collected from Okha.

Description: This species inhabits the Soft sediment, where an anemone lives with the column commonly buried in the substratum with oral disc visible at surface. Oral disc is flat and can be up to 500mm diameter when fully expanded. It is usually brownish or grey, rarely bright green, sometimes with white stripes. The column is widening gradually from the pedal disc and flared at oral end. The column is generally whitish, violet or greenish with large, prominent, adhesive bumpy verrucae. Texture is leathery and each prominent verrucae with raised rim. Tentacles are sinuous and can be up to 45mm long, they are so long that they typically become intertwined. Tentacles are all over the surface of the oral disc, giving it a 'mop' appearance. They can be a brownish-gray, mauve or green; and some specimens are found with beautiful blue, purple, and mauve tips. The mouth is elongated. Base is adherent, generally flat and rarely exceeding diameter of lower column. Sphincter

mussels diffuse to circumscribed, usually not very strong. Retractor muscles are weaker and diffuse (Plate: 4.20).

Distribution: Previously recorded from Singapore, Pulau Hantu (England, 1987), shallow tropical and subtropical seas from the Red sea, across the Indian ocean, to New Caledonia and Japan to Australia. ((Dunn, 1981, Fautin and allen, 1992, Fautin, 2008). North, Middle, South and Little Andaman, MGMNP, Ritchie's Archipelago and Nicobar (Madhu and Madhu, 2007, Raghunathan *et al.*, 2014). Mandapam - Southeast coast of India. (Subramanian *et al.*, 2011). We have reported this species from the Okha and Mithapur coast.

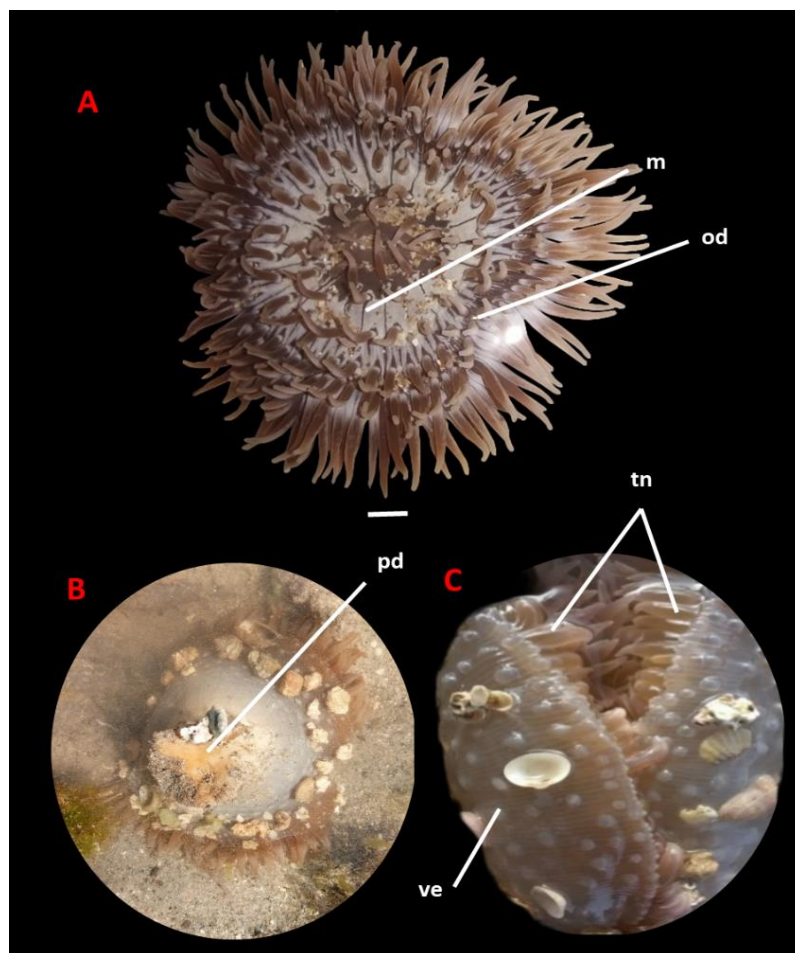


Plate 4.19: *Heteractis crispa* (Ehrenberg, 1834). A: Oral view of live specimen B: Aboral view C: Column details Mouth (m), Verrucae (ve), Pedal disc (pd), Oral disc (od), Tentacles (tn). Scale bar: 10mm

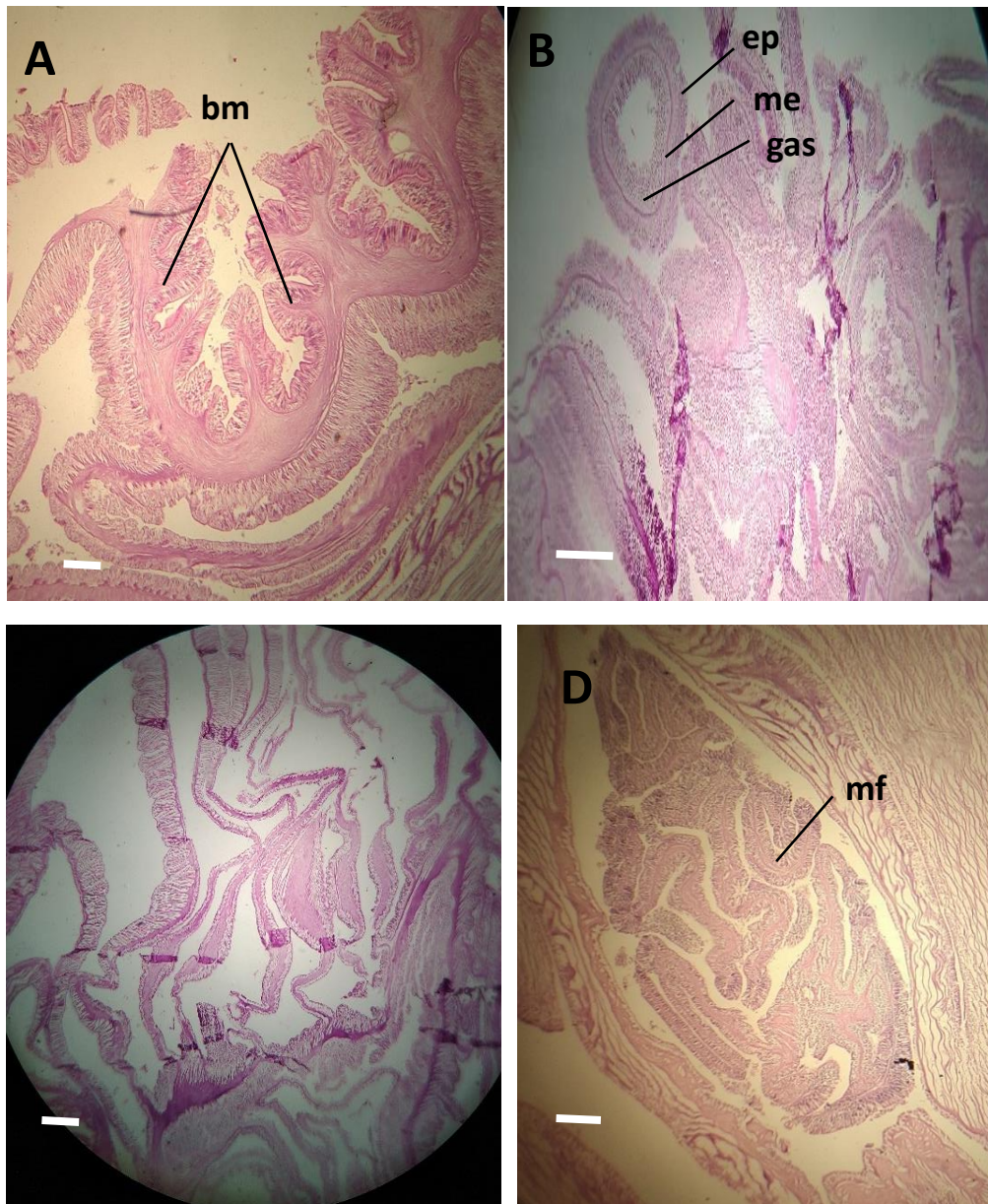


Plate 4.20: Histology of *Heteractis crispa* (Ehrenberg, 1834).

A: Longitudinal section through base showing basilar muscles **B:** Longitudinal section through upper column **C:** Longitudinal section through margin **D:** Cross section showing mesenterial filaments. **bm:** basilar muscle **ep:** epidermis **me:** mesoglea **gas:** gastrodermis **mf:** mesenterial filaments. Scale bar: 0.1mm

15. *Heteractis magnifica* (Quoy and Gaimard, 1833) (Plate: 4.21)

Synonyms:

Actinia magnifica (de Blainville, 1830)
Actinia magnifica (Quoy & Gaimard, 1833)
Antheopsis ritteri (Stephenson, 1922)
Corynactis magnifica (Milne Edwards, 1857)
Helianthopsis mabrucki (Carlgren, 1900)
Helianthopsis ritteri (Kwietniewski, 1898)
Heteractis ritteri (Cutress & Arneson, 1987)
Radianthus mabrucki (Stephenson, 1922)
Radianthus paumotensis (Friese, 1972)
Radianthus ritteri (Carlgren, 1949)
Ropalactis magnifica (Andres, 1883)
Radianthus malu (Allen, 1978)

Common Name: Ritteri Anemone, Magnificent Anemone

Material Examined: OKHHM01 (22° 28' 16.3" N, 69° 4' 33.5" E) (CH-51mm, CD-28mm, LD-30mm, PDD-33mm, ODD-98mm) collected from Okha.

Description: This species is mostly sub tidal but may be exposed at unusually low tides. They occupy fully exposed, prominent position attached to the solid object such as coral boulder. Oral disc is flat to gently undulating, brownish and may exceed 500mm diameter, but typically less than that. Central oral disc is yellow, brown or green and often raised so that mouth sits on a cone. Cylindrical column is thin, somewhat smaller diameter than oral disc, equal to pedal disc of uniform bright colour Commonly like Red, Blue, Green, Yellow and Magenta with distal verrucae. Column with longitudinal rows of translucent verrucae same colour as column or slightly lighter or darker or slightly lighter or darker.

Distribution: Previously recorded from Singapore (England, 1987), shallow tropical and subtropical seas from the Red sea, cross the Indian ocean, to New Caledonia and Japan to Australia. ((Dunn, 1981, Fautin and allen, 1992, Fautin,

2008). North, Middle and Little Andaman, MGMNP, Ritchie's Archipelago and Nicobar (Madhu and Madhu, 2007, Raghunathan *et al.*, 2014). We have been recorded this species from the Okha and Mithapur coast of Gujarat coast.

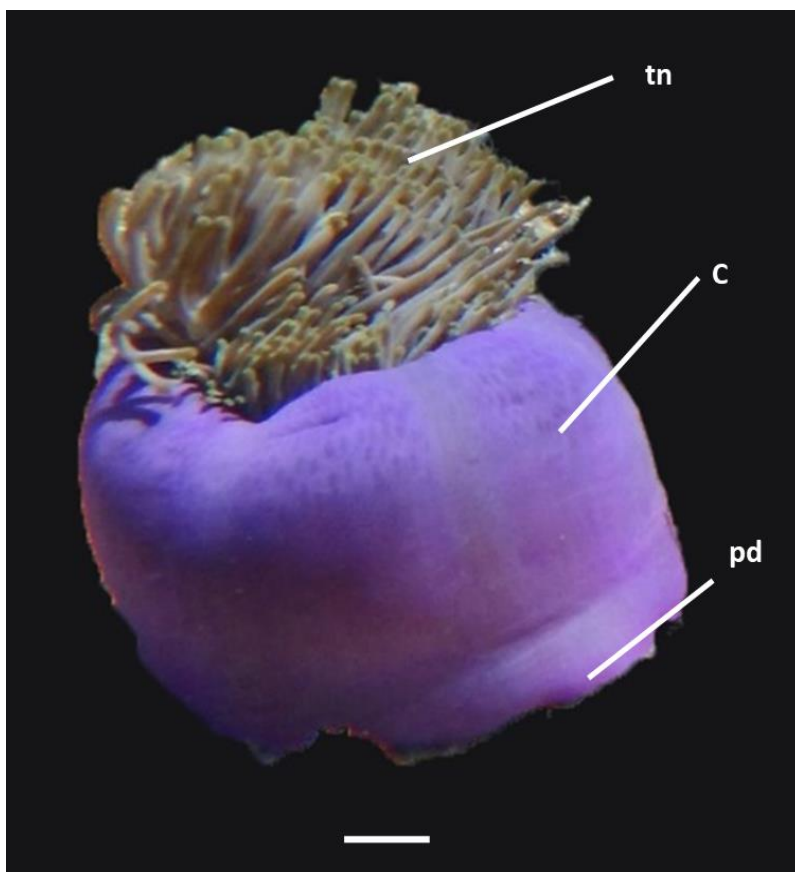


Plate 4.21: *Heteractis magnifica* (Haddon and Shackleton, 1893). A, B: Oral view of living specimen C: Tentacle details. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

16. *Heteractis malu* (Haddon and Shackleton, 1893) (Plate: 4.22)

Synonyms:

Discosoma Malu (Haddon & Shackleton, 1893)

Stichodactis papillosa (Kwietniewski, 1898)

Antheopsis concinnata (Lager, 1911)

Stichodactis glandulosa (Lager, 1911)

Macranthea cookie (Verrill, 1928)

Antheopsis malu (Haddon & Shackleton, 1893)

Antheopsis papillosa (Kwietniewski, 1898)

Radianthus malu (Haddon & Shackleton, 1893)

Radianthus papillosa (Kwietniewski, 1897)

Common name: Delicate sea anemone

Material Examined: OKHHMU01 (22° 28' 42.66"N, 69° 4' 48.69"E) (CH-30mm, CD-26mm, LD-27mm, PDD-29mm, ODD-108mm) collected from Okha

Description: They are found in sandy and gravel areas mostly burrowed in the soft sediment. Oral disc lies at surface of sediment in which delicate column is burrowed. It is brown, brown, and purple, green with white markings and can be up to 200mm diameter. Mouth is 10-20mm long and slit like to circular. Tentacles sparse, stubby (rarely to 40 mm long), of variable length even within one radial row, commonly magenta-tipped. Tentacles arise from brown or purplish (rarely bright green) oral disc may have white radial markings; evenly tapered to point or slightly inflated in middle; lower part same colour as oral disc, but upper portion may have several white rings or green end. Column very thin in expansion; upper part violet-brown (due to zooxanthellae) with longitudinal rows of adhesive verrucae. Column commonly pale cream or yellow colour, may have splotches of deep yellow or orange.

Distribution: Indonesia; Papua New Guinea; Japan; Hawaii; Australia from Broome to Point Peron in Western Australia, Torres Strait to Moreton Bay in Queensland (Fautin *et al.*, 2008). Also reported from South, Middle, and North Andaman (Raghunathan *et al.*, 2014). We have reported from Okha.

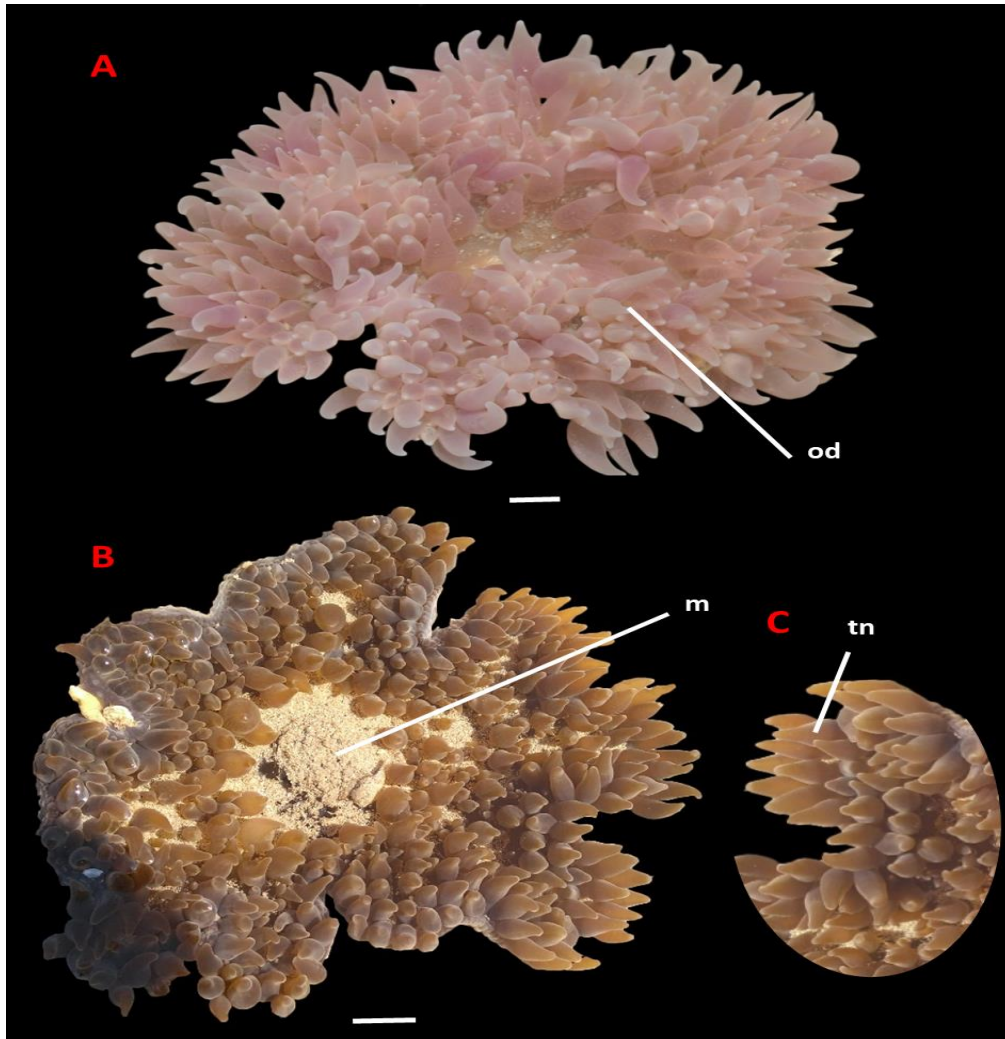


Plate 4.22: *Heteractis malu* (Haddon and Shackleton, 1893). A,B: Oral view of living specimen C: Tentacle details. Mouth (m), Oral disc (od), Tentacles (tn). Scale bar: 10mm

Genus *Stichodactyla* (Brandt, 1835)

Pedal disc well developed, adherent. Flat, undulating, and broad oral disc covered with many short tentacles; endocoelic tentacles radially arrayed, mostly two or more abreast, to five at margin; rows communicating with successively higher order endocoels shorter, more marginal; in most species all tentacles alike, but single exocoelic ones may be more robust. Column generally broader than tall, flared part verrucose. Fosse absent to shallow. Cnidom: spirocysts, basitrichs, micro basic p-mastigophores. (Fautin *et al.*, 2008).

17. *Stichodactyla gigantea* (Forskal, 1775) (Plate: 4.23)

Synonyms:

Actinia amethystina (Quoy & Gaimard, 1833)

Actinia gigantea (Forskal, 1775)

Actinia parvitentaculata (Quoy & Gaimard, 1833)

Discosoma kenti (Haddon & Shackleton, 1893)

Polyparium ambulans (Korotneff, 1886)

Priapus giganteus (Forsskal, 1775)

Radianthus parvitentaculatus (Quoy & Gaimard, 1833)

Stichodactyla kenti (Haddon & Shackleton, 1893)

Stoichactis gigantea (Forsskal, 1775)

Stoichactis giganteum (Forsskal, 1775)

Stoichactis giganteus (Forsskal, 1775)

Stoichactis intermedia (Lager, 1911)

Stoichactis kenti (Haddon & Shackleton, 1893)

Common name: Merten's carpet anemone

Material examined: OKHSG01 (22° 28' 46.823" N, 69° 4' 49.79"E) (CH-32mm, CD-26mm, LD-28mm, PDD-32mm, ODD-98mm) collected from Okha.

Description: This anemone inhabits sandy shallow water and is attached to rock buried in sand. This anemone can be sticky to touch and tentacles that adhere to a human finger can pull off the animal. Oral disc rarely as much as 500 mm diameters. Oral disc is thick, broad, smooth and highly folded. Oral disc around mouth bare but mouth may be hidden amidst folds of oral disc. Mouth is grey in colour. Verrucae are present on upper column non-adhesive, blue to maroon, contrasting with yellowish, pinkish, tan, greenish-blue, or gray-green column. Column considerably narrower, relatively short. Tentacles are numerous, small, sturdy, evenly distributed, extremely sticky, tip slightly tapered. They are about 8 mm long and move constantly, making this anemone look like its vibrating.

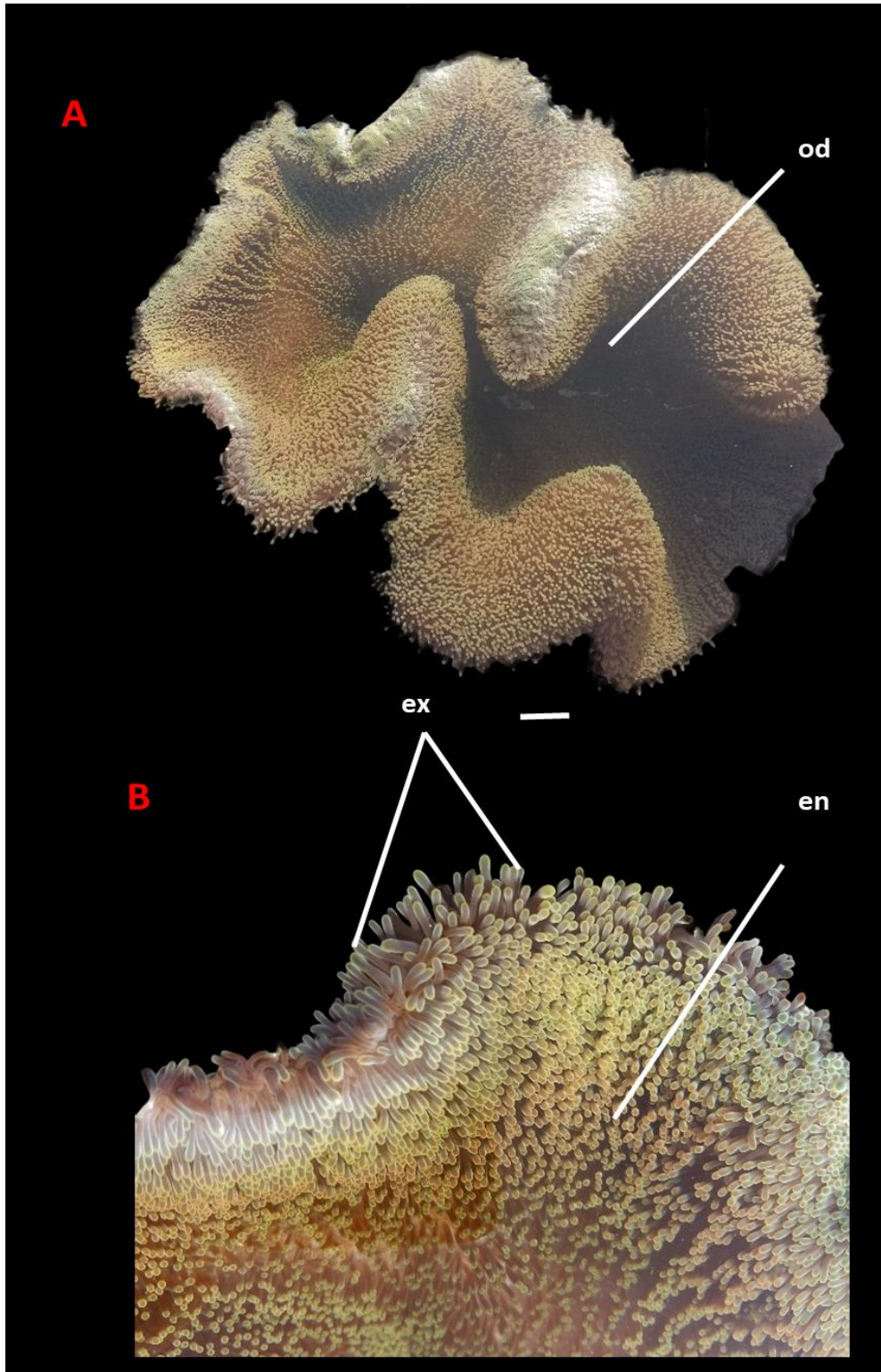


Plate 4.23 *Stichodactyla gigantea* (Forsk., 1775). **A:** Oral view of living specimen **B:** Tentacle details. Oral disc (od), exocoelic tentacles (ex), endocoelic tentacles (en). Scale bar: 20mm

Distribution: Previously recorded in Singapore from Pasir Panjang, Buona Vista, Bedok, Pulau Hantu and Cyrene Reef by England (1987) who reported ad *S. kenti*. Also occurs in shallow tropical and subtropical seas from the Red sea, across the Indian Ocean, to New Caledonia and Japan to Australia. (Dunn, 1981; Fautin and allen, 1992; Fautin, 2008). From India, Gulf of Mannar, and Andaman and Nicobar Islands Raghunathan *et al.*, 2014). We have reported distribution of this species from Okha.

18. *Stichodactyla haddoni* (Saville-Kent, 1893) (Plate: 4.24)

Synonyms:

Stoichactis haddoni (Saville-Kent, 1893)

Stoichactis amboinensis (Pax, 1924)

Discosoma haddoni (Saville-Kent, 1893)

Common Name: Haddon's Carpet Anemone, Saddle carpet anemone

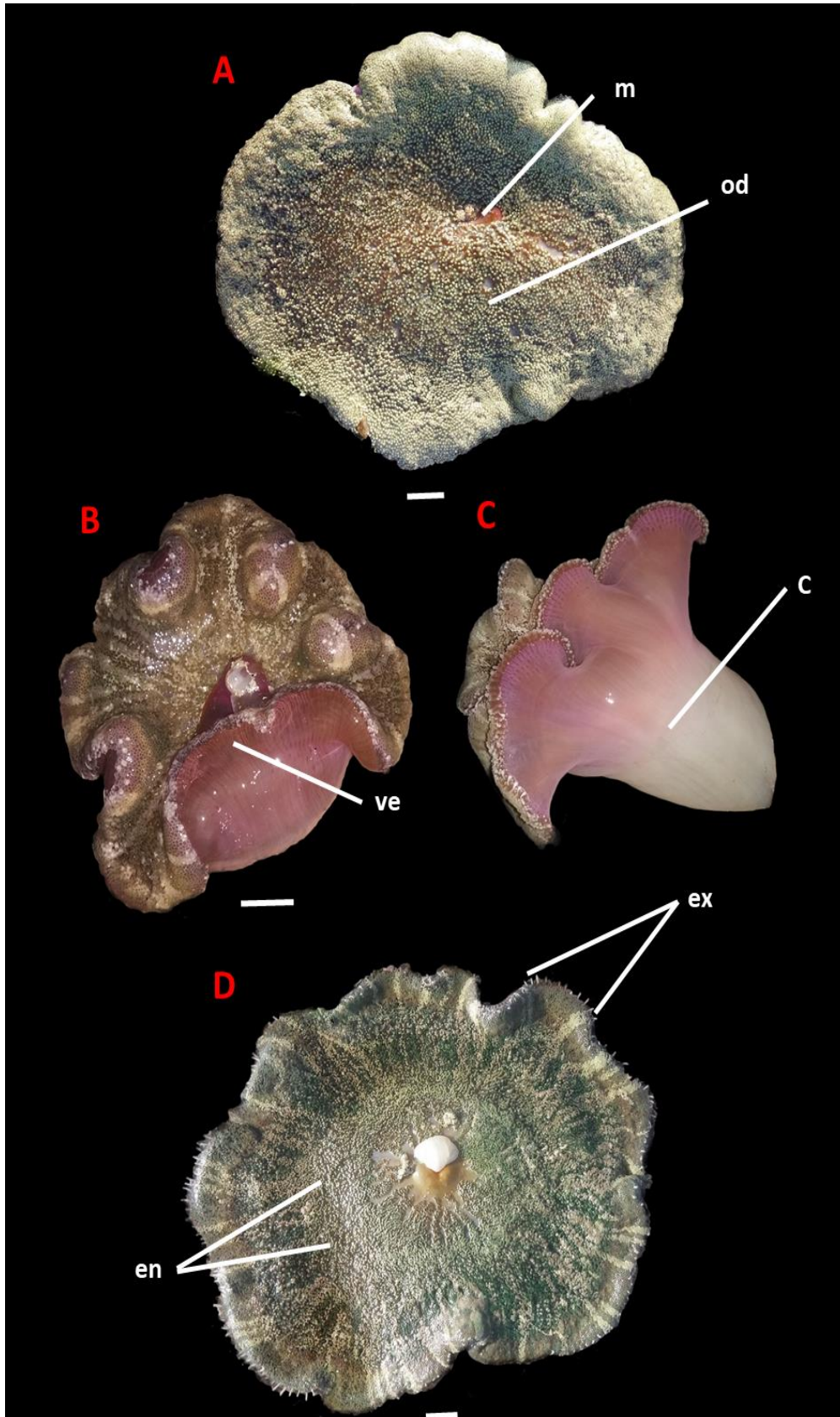
Material Examined- OKHSH01 (22° 28' 50.79" N, 69° 4' 48.17" E) (CH-30mm, CD-27mm, LD-29mm, PDD-35mm, ODD-130mm), OKHSH02 (22° 28' 40.8" N, 69° 5' 48.36" E) (CH-20mm, CD-18mm, LD-19mm, PDD-28mm, ODD-180mm) OKHSH03 (22° 28' 32.18" N, 69° 4' 28.15" E) (CH-32mm, CD-28mm, LD-31mm, PDD-39mm, ODD-160mm) OKHSH04 (22° 28' 32.18" N, 69° 4' 48.17" E) (CH-43mm, CD-38mm, LD-40mm, PDD-44mm, ODD-210mm) collected from the Okha coast.

Description: Saddle anemones may found in areas where the column may burrow into soft sediments from mid tide to as deep as adequate light penetrates. They are often found among seagrasses in shallow waters, but are also found to be attached with rocks or coral substrates. The form is also seen in other colours. It may be green, opaque green, or dark brown with light green or white stripes at the oral disc's margin. Oral disk diameter, when expanded, is usually 250-500 mm. *S. haddoni* has two types of tentacles. (1) Exocoelic tentacles, pointed white tentacles longer than endocoelic tentacles; they are only present at the edge of the oral disc, and each alternate with many rows

of small spherical tentacles on the oral disc (2) Endocoelic tentacles, internal; each group of white tentacles emerges simultaneously and is the same size or shorter than exocoelic tentacles; they are distinct in all parts of the oral disc. Each exocoelic tentacle of approximately 5 mm in length alternates on the oral disc with rows of short spherical endocoelic tentacles. The Endocoelic tentacles are dark brown and opaque green with gray and purple colouration in some parts. Not all tentacles are alike, and near the mouth their density is lower. There are no tentacles around the mouth about 12 mm.

Oral disc is undulated and wider than pedal disc. Mouth is red and pedal disc is white to yellow or light brown. Column commonly whitish or brownish with rose or purple nonadhesive verrucae tappers to pedal disc much narrower than oral disc. Rows of nonadhesive verrucae are visible on the column. The *S. haddoni* have not been bred in captivity and rarely do they split on their own.

Distribution: *Stichodactyla haddoni* occurs throughout the Indian Ocean and the central Indo-West Pacific, ranging to the west from the Red Sea and the Gulf of Oman southward along the East African coast to southern Mozambique (Inhaca Island, ca. 26°S), and to the east from the Izu and Ryukyu Islands (Japan) southward to the Solomons Islands, the Great Barrier Reef, New Caledonia and the Fiji Islands (Dunn, 1981; Fautin and Allen, 1992; Fautin, 2008). From India, it has been reported from Off Tutticorin, Mandapam coast (Thangaraj *et al.*, 2011) and Andaman Nicobar island (North, Middle and South) (Raghunathan *et al.*, 2014). From Gujarat, it has been recorded previously from Gulf of Kachchh, Gulf of Mannar, Mithapur and Okha (Trivedi 1977, Nayar and Mahadevan, 1967, Hartog and Vennam, 1993). We have reported distribution of this species from Okha and Mithapur.



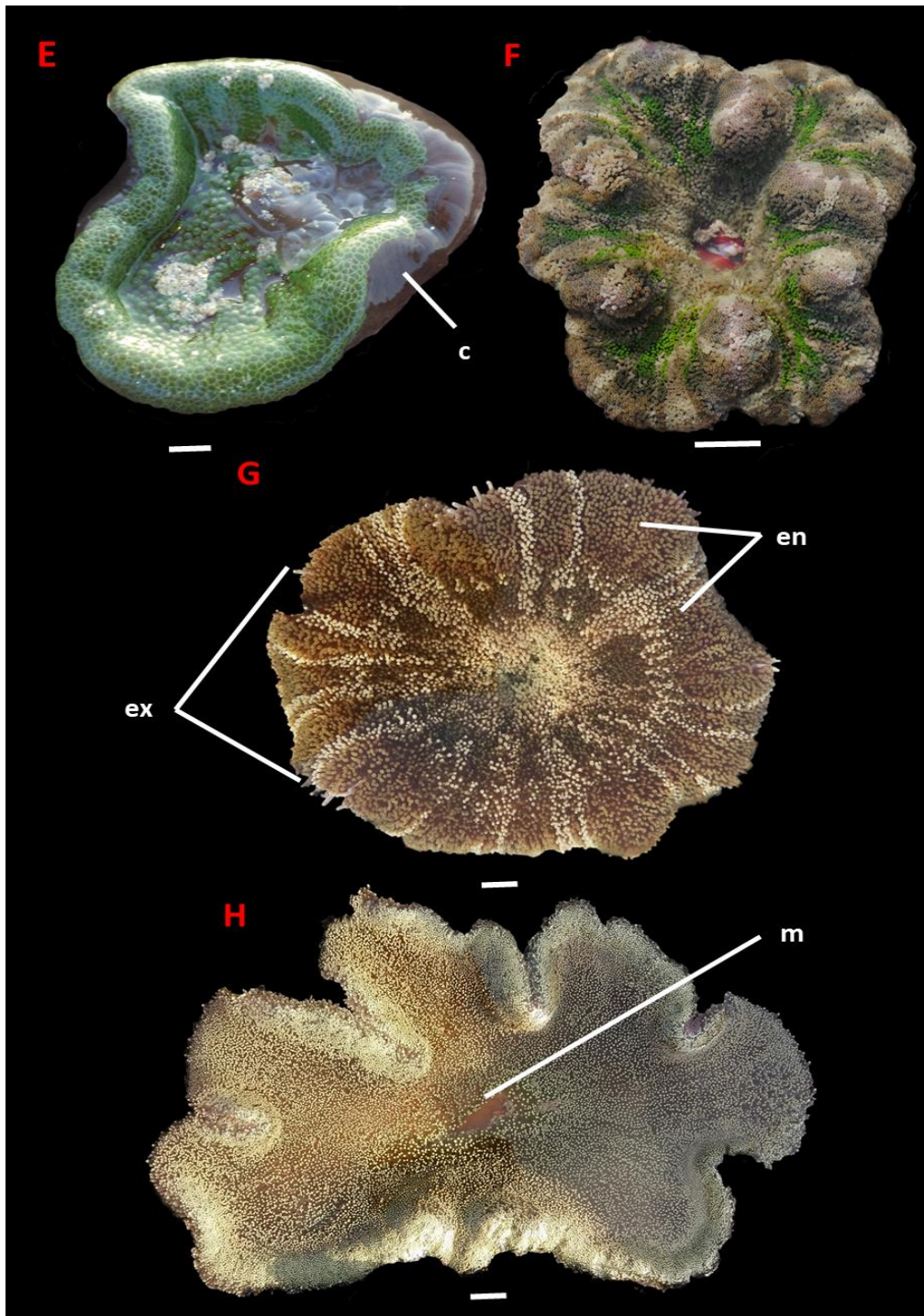


Plate 4.24: *Stichodactyla haddoni* (Saville-Kent, 1893). A: Oral view of live specimen B, C: Column details D: Tentacle details E, F, G, and H: Morphs of specimen. Mouth (m), Verrucae (ve), Pedal disc (pd), Oral disc (od), exocoelic tentacles (ex), endocoelic tentacles (en). Scale bar: 20mm

19. *Stichodactyla mertensii* (Brandt, 1835)(Plate: 4.25)

Synonym:

Stoichactis giganteum (Mariscal, Allen and Mariscal 1971, Allen 1972, 1973, 1975)

Common name: Merten's carpet anemone

Material examined: OKHSM01 (22° 28' 31.19"N, 69° 4' 50.74"E) (CH-19mm, CD-13mm, LD-15mm, PDD-17mm, ODD-86mm) collected from the Okha coast of Saurashtra.

Description- This species mostly live attached to coral rubble and rocky walls and crevices, hiding most of its body with only oral disc remaining above the substrate. This is the smallest species of this genus compared to other species. The oral disc shape is often more ovoid than circular. Oral disc are flared, thick, slightly undulated, pale in colour. Oral disc tan to white column with longitudinal rows of verrucae pigmented magenta or orange. These adhesive verrucae, being under the oral disc edge, hold the disc in position. No verrucae below wide upper column, but splotches of pigment continue down short or less longitudinal streaks. Tentacles are in two types, short tentacles evenly distributed on the oral disc, which is greenish orange in colour and the long ones are pale or white in colour, which are radially arranged from center of the oral disc to periphery at even distance. Tentacles are bubble in shape- all may be short (10-20 mm long), or some (in patches) very long (to 50 mm or more). Tan oral disc almost entirely covered with tentacles; yellow or greenish tentacle-free oral area 20-50 mm diameter.

Distribution: Micronesia and Melanesia to East Africa, and Australia to the Ryukyu Islands, Madang province (Fautin, 1988). From India, it has been recorded from Andaman and Nicobar island (North) (Raghunathan *et al.*, 2014). We have reported this species from the Okha site of Saurashtra coast, Gujarat.

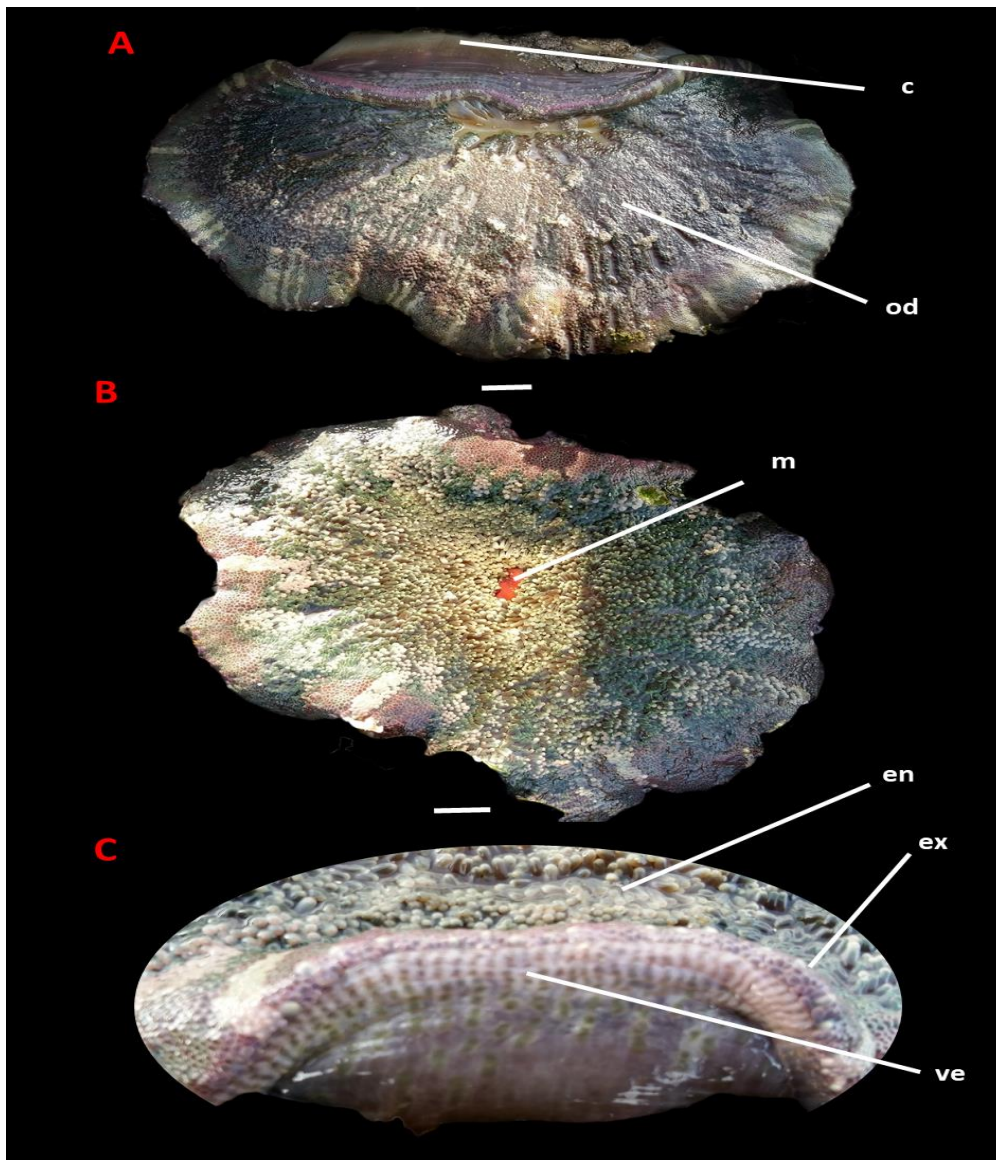


Plate 4.25: *Stichodactyla mertensii* Brandt, 1835. A, B: Oral view of living specimen C: Tentacle and column details. Mouth (m), Verrucae (ve), Oral disc (od), exocoelic tentacles (ex), endocoelic tentacles (en). Scale bar: 20mm

20. *Stichodactyla tapetum* (Hemprich & Ehrenberg in Ehrenberg, 1834)(Plate:26)

Synonyms:

Actinia (Isacmaea) Tapetum (Hemprich & Ehrenberg in Ehrenberg, 1834)

Stoichactis laevis (Lager, 1911)

Homactis rupicola (Verrill, 1879)

Discosoma amboinensis (Kwietniewski, 1898)

Stoichactis tapetum (Carlgren, 1900)

Stoichactis australis (Lager, 1911)

Stoichactis rupicola (Carlgren, 1949)

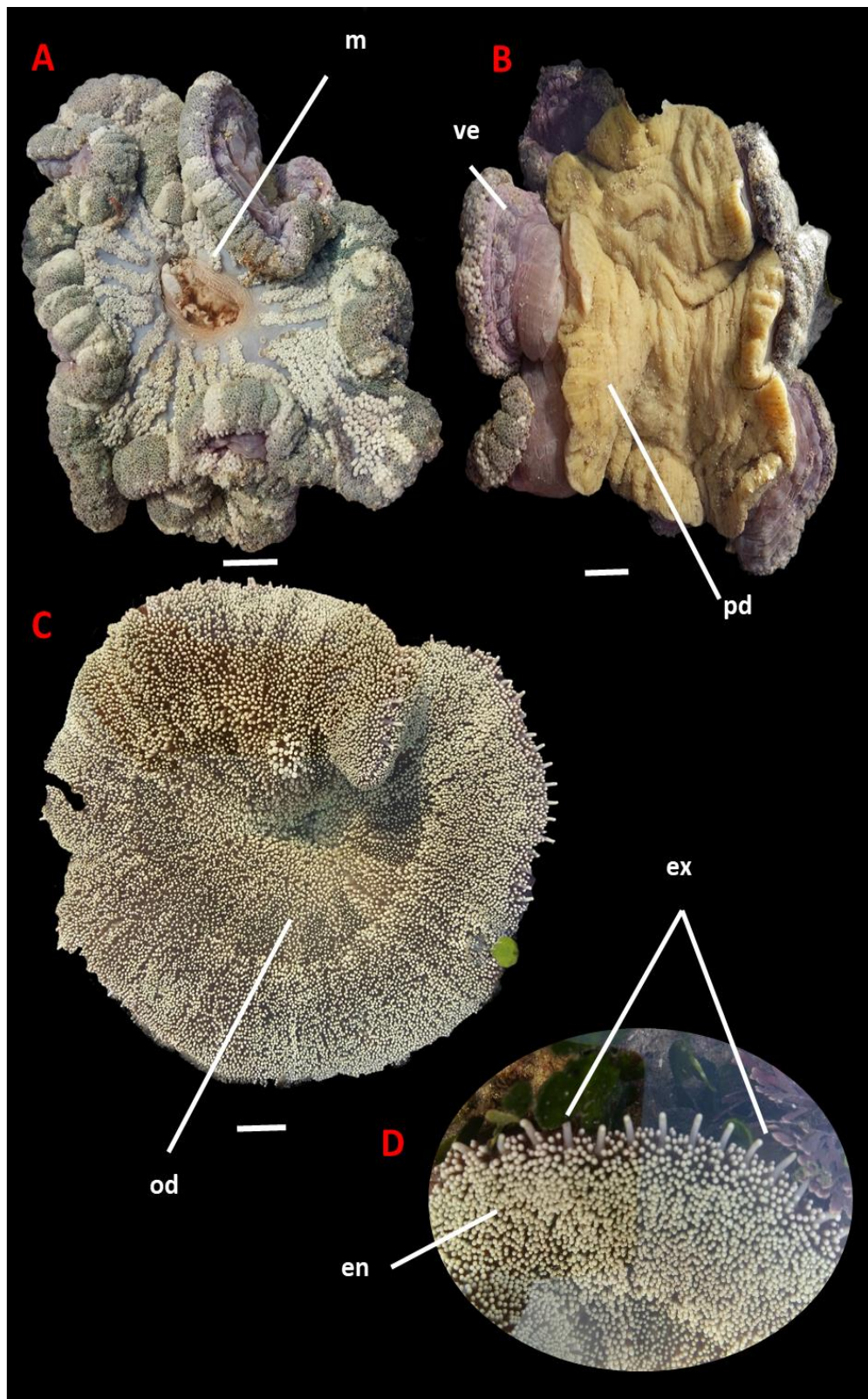
Common name: Mini carpet anemone

Material examined: OKHST01 (22° 28' 46.35" N, 69° 4' 48.67" E) (CH-18mm, CD-12mm, LD-17mm, PDD-20mm, ODD-80mm), OKHST 02 (22° 29' N, 69° 05' E) (CH-20mm, CD-16mm, LD-17mm, PDD-19mm, ODD-90mm) collected from Okha.

Description: This species inhabits in crevices of rocks and pebbles, on mud and sand flats. Tentacles that are very thin, bulbous, not longer than 1 mm, less than 0.5 mm in diameter, densely packed with a long rectangular axis perpendicular to the radius. Tentacles rectangular up to 1 mm across a long axis. Those who communicate with a single endocoel can vary in size due to crowding at least two abreast, often as many as six across. The marginal colonial tentacles, in particular, are 1.5-2.0 mm long. Tentacles are green, all or only exocoelic ones, with white tips. Oral disc is flat and occasionally with lips. Oral disc diameter of widely spread specimen can be up to 100-150 mm. Rows of tentacles communicating with first or first and second order endocoel approach mouth to even reach it. Tentacles communicating with endocoel in wedge-shaped row, with mostly five obvious cycles. Tentacles communicating with each endocoel in wedge shaped row. Column smooth (no verrucae on column), texture firm due to thick mesoglea, narrower diameter towards middle part from pedal disc, flaring abruptly at distal end. Pedal disc and column red to dark pink with narrow white strips. Column length typically about one-half pedal disc diameter, but may be taller. Sex is separate.

Distribution: From Japan to Australia, Indian Ocean to New Caledonia and Red Sea (Dunn, 1981; Fautin and Allen, 1992; Fautin, 2008), and Singapore (Fautin

et al., 2009); south-eastern coast of Iran (Fariman and Javid, 2015). However, from India it has been recorded for the first time from Gujarat.



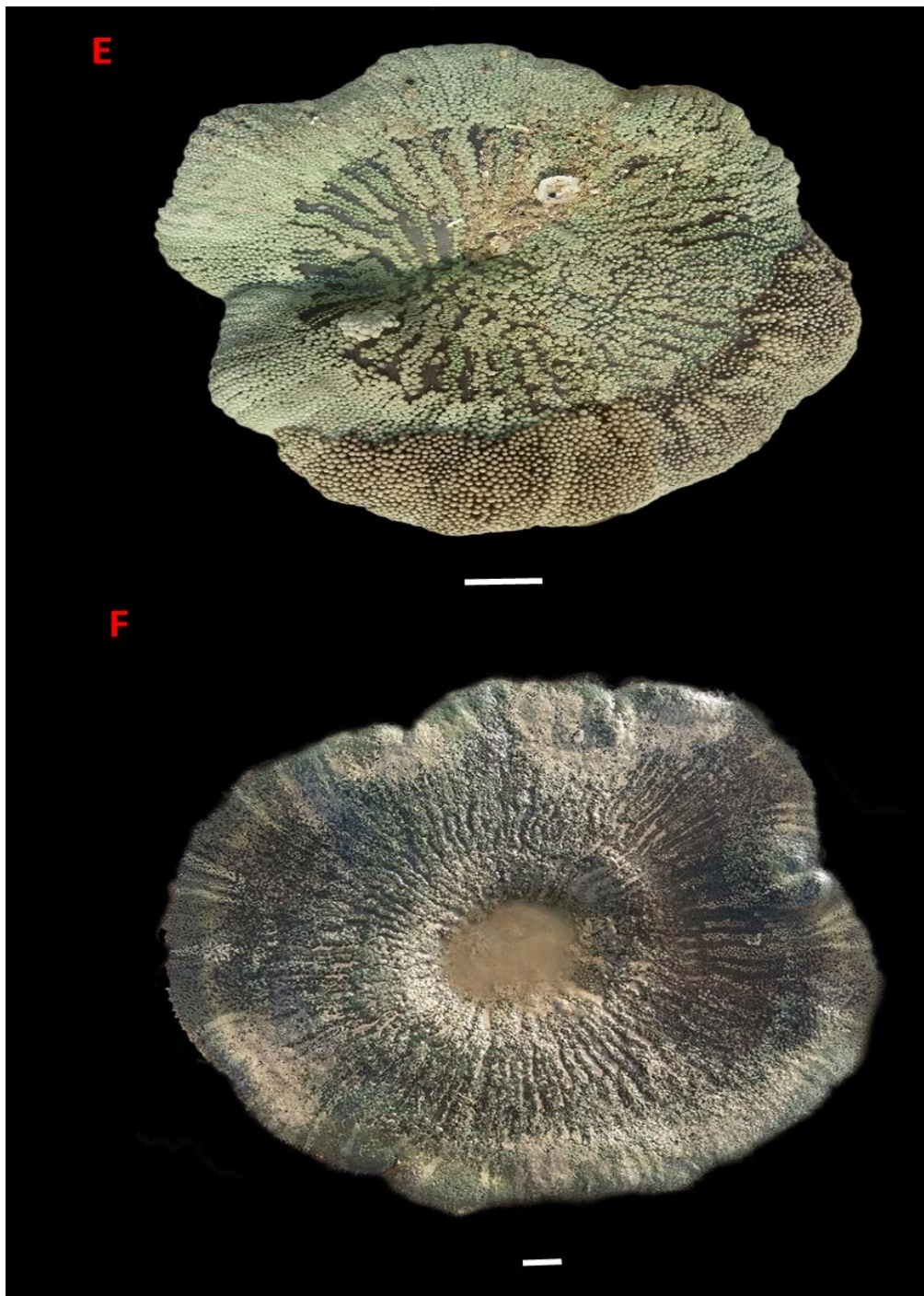


Plate 4.26: *Stichodactyla tapetum* (Hemprich & Ehrenberg in Ehrenberg, 1834).

A: Contracted live specimen B: Aboral view of living specimen C: Oral view of living specimen D: Tentacle details E, F: Morphs of specimen. Mouth (m), Verrucae (ve); Pedal disc (pd), Oral disc (od), exocoelic tentacles (ex), endocoelic tentacles (en). Scale bar: 20mm

Family: Thalassianthidae (Milne Edwards, 1857)

Thenaria with a well-developed base (Endomyaria). Column in the upper part with more or less distinct verrucae. Sphincter weak, restricted or circumscribed. Oral disks often thrown into various permanent lobes that are small, cyclically arranged; or sometimes not. The lobes, if present, hold a group of nematospheres on the oral side of the dendritic tentacles that are continued on the disk and radially arranged. A loop of exocoelic dendritic tentacles at the top. Parietobasilar muscles weak, basilar muscles well developed (Carlagen, 1949).

Genus *Cryptodendrum* (Klunzinger, 1877)

Cryptodendrum genus is monotypic with a single species. Thalassianthidae with a wide pedal disk. Small verrucae. Wide, irregularly folded oral disc at the top of the column. Two kinds of tentacles, often short sometimes dendritic, sometimes spherical. There is a single row of dendritic, exocoelic tentacles at the bottom, inside which there is a continuous, broad band of globular nematosphere. Apart from directives, all stronger mesenteries are fertile. At the top, more mesenteries than at the bottom. Diffusing, Retractors diffuse, well-developed, band-like. Parietobasilar muscles weak, basilar muscles distinct (Carlagen, 1949).

21. *Cryptodendrum adhaesivum* (Milne Edwards, 1857)(Plate:27)

Synonyms:

Cryptodendrum adhasivum (Klunzinger, 1877)

Stoichactis digitate (Doumenc, 1973)

Common Name: Pizza Anemone, Sticky Carpet Anemone, Nap-Edged Anemone

Material examined: OKHCA01 (22° 28' 31.82" N, 69° 4' 49.65"E) (CH-29 mm, CD-32mm, LD-34mm, PDD-37mm, ODD-116mm) collected from the Okha coast.

Description: Mostly found on crevices in shallow water or coral rubble. They are spreading over or between the rocks. Usually only broad oral disc is visible. Oral disc can reach up to 200-300 mm diameter. The mouth is low (about 10 mm) in diameter at the middle of the oral disk. It is generally a contrasting colour to the disk, like yellow, white green or violet. The pedal column of anemone has sticky foot which adheres to the various surface. The pedal column has white, orange or yellow coloured row of verrucae or small bulges. They may also have lines, flicks and spots on it. The column of the body is brightly coloured; generally smooth with tiny verrucae of the same or slightly darker colour. Extremely sticky tentacles; short (to 5 mm long), thick, of two forms: those in the centre of the oral disk have a narrow stalk with five or more short branches at the end (i.e. like a miniature glove); those near the edge are simple elongated bulbs about 1 mm in diameter; at the extreme periphery is a ring of tentacles like the central ones but with fewer branches. Tentacles of the two types typically vary in colour: found variations include yellow and pink blue and gray green and brown; often tentacles of a different colour appear in patches between those of predominant colour. Tentacle stalk and tips can also vary in colour, so it can be highly colourful. No sexual difference in appearance is known in this anemone.

Distribution: Occurs throughout the tropical and subtropical Indo West Pacific in shallow water, from the Red Sea, across the Indian Ocean, to French Polynesia, and Japan to Australia (Dunn, 1981; Fautin and Allen, 1992; Fautin, 2008). Also reported from South, Middle, and North Andaman (Raghunathan *et al.*, 2014). We documented this species from Okha.

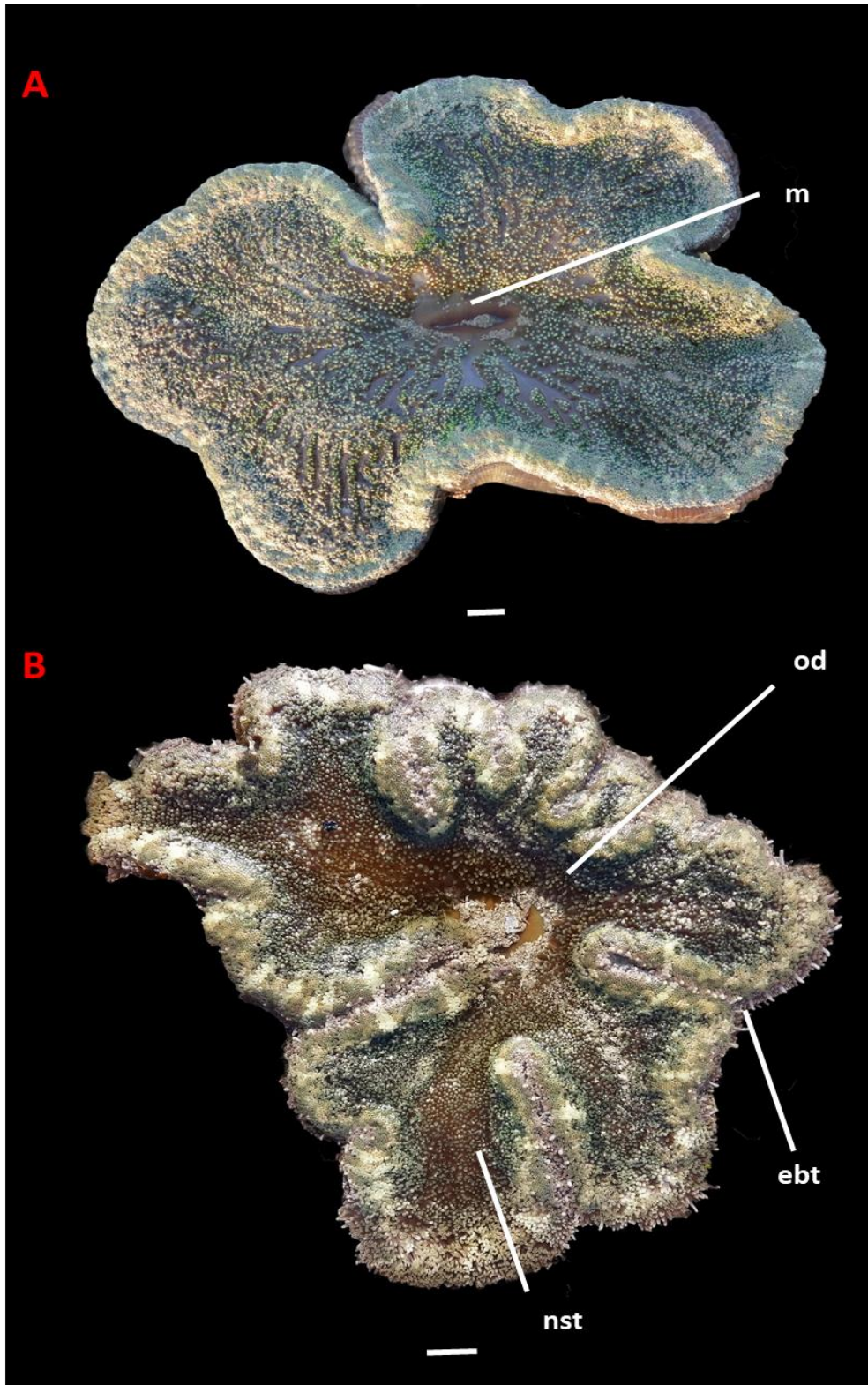


Plate 4.27: *Cryptodendrum adhaesivum* Milne Edwards, 1857. A, B: Oral view of living specimen. Mouth (m), Oral disc (od), Narrow stalk tentacles (nst), elongated bulbous tentacles (ebt). Scale bar: 20mm

4.2 MOLECULAR IDENTIFICATION OF SEA ANEMONES

DNA isolation and quantification

Total 25 samples of DNA were successfully isolated out of 35 samples. Samples of DNA could be successfully isolated as evidenced by Gel profile here in (Fig. 4.1). Inability to isolate DNA is attributable to high salt contents that required modifications of standard method. Results of DNA yield extracted from sea anemones are shown in figure. It shows different concentration of DNA obtained from all the samples of sea anemone with very bright band indicates very high yield of good quality DNA, while the faded bands indicate low yields and fragmented DNA content. Samples showing faded bands were quantified using QI Expert and the concentration of DNA at 260 above 1.6-1.8 was taken for amplification.

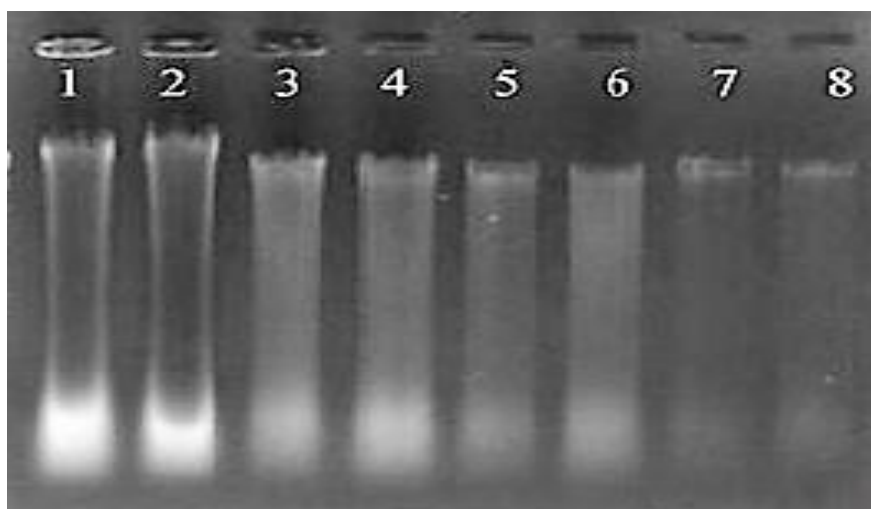


Figure 4. 1 : Gel image of DNA isolation

PCR amplifications

The DNA isolated from sea anemone samples were amplified using NS1 and NS4 primers. The amplified product was confirmed by observing on agarose gel through gel electrophoresis (Fig. 4.2). Samples containing positive results were further processed for PCR purification and cycle sequencing steps.

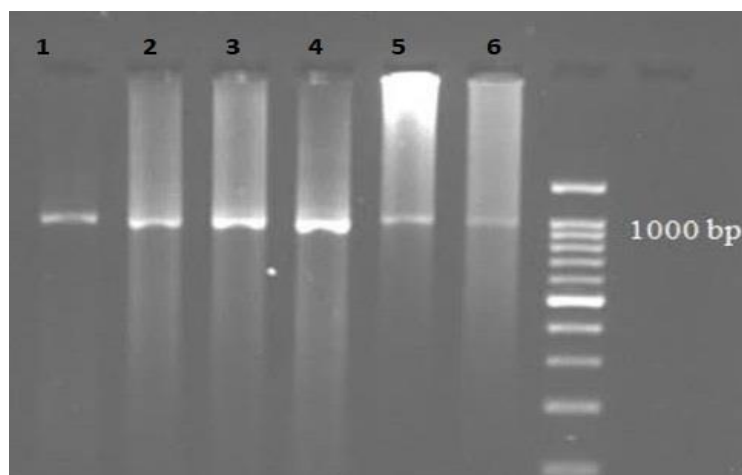


Figure 4. 2 : Gel image of PCR amplification

Sequencing results

DNA sequences of each species were separately aligned using Bio Edit 7.2 software using the following settings and parameters: Strategy, L-INS-I (recommended for, 18 sequences with one conserved domain and long gaps); scoring matrix, 200PAM/k $\frac{1}{4}$ 2; gap opening penalty, 1.53; offset value, 0.05; max. Iterate, 1000; and retree, 1. We then concatenated the nuclear marker sequences to create a dataset for sea anemone species. A total of 18 sea anemone sequences of 18S rDNA were acquired from different morphologically identified species from the Saurashtra coastal zone. The sequences were compared to worldwide available Genbank sequences data for Sea anemone fauna and Genbank accession number were obtained (Table: 4.1). Phylogenetic frames can disclose the evolutionary patterns of many morphological characteristics. The tree using Maximum likelihood method shows the intraspecific and interspecific relatedness amongst the same species, found at different locations of the world.

| Species | Species Name | Locality | Accession No. |
|---------|----------------------------------|---------------|---------------|
| 1 | <i>Actinia equine</i> | Sutrapada | MN905020 |
| 2 | <i>Anemonia viridis</i> | Shivrajpur | MN905048 |
| 3 | <i>Anthopleura anjunae</i> | Vadodra jhala | MN907372 |
| 4 | <i>Anthopleura artemisia</i> | Sutrapada | MN508442 |
| 5 | <i>Anthopleura elegantissima</i> | Vadodra jhala | MN918266 |
| 6 | <i>Anthopleura sola</i> | Vadodra jhala | MN905046 |
| 7 | <i>Urticina clandenstina</i> | Vadodra jhala | MN519729 |
| 8 | <i>Exaiptasia diaphana</i> | Shivrajpur | MN905026 |
| 9 | <i>Diadumene lineata</i> | Sutrapada | MN918282 |
| 10 | <i>Phymanthus crucifer</i> | Okha | MN922527 |
| 11 | <i>Heteractis crista</i> | Okha | MN918283 |
| 12 | <i>Heteractis magnifica</i> | Okha | MN515148 |
| 13 | <i>Heteractis malu</i> | Okha | MN918272 |
| 14 | <i>Stichodactyla gigantea</i> | Okha | MN518343 |
| 15 | <i>Stichodactyla haddoni</i> | Okha | MN905027 |
| 16 | <i>Stichodactyla mertensii</i> | Okha | MN515165 |
| 17 | <i>Stichodactyla tapetum</i> | Okha | MN905022 |
| 18 | <i>Cryptodendrum adhaesivum</i> | Okha | MN907373 |

Table 4. 1: Sea anemone taxa included in the study with locality and Genebank Accession Number

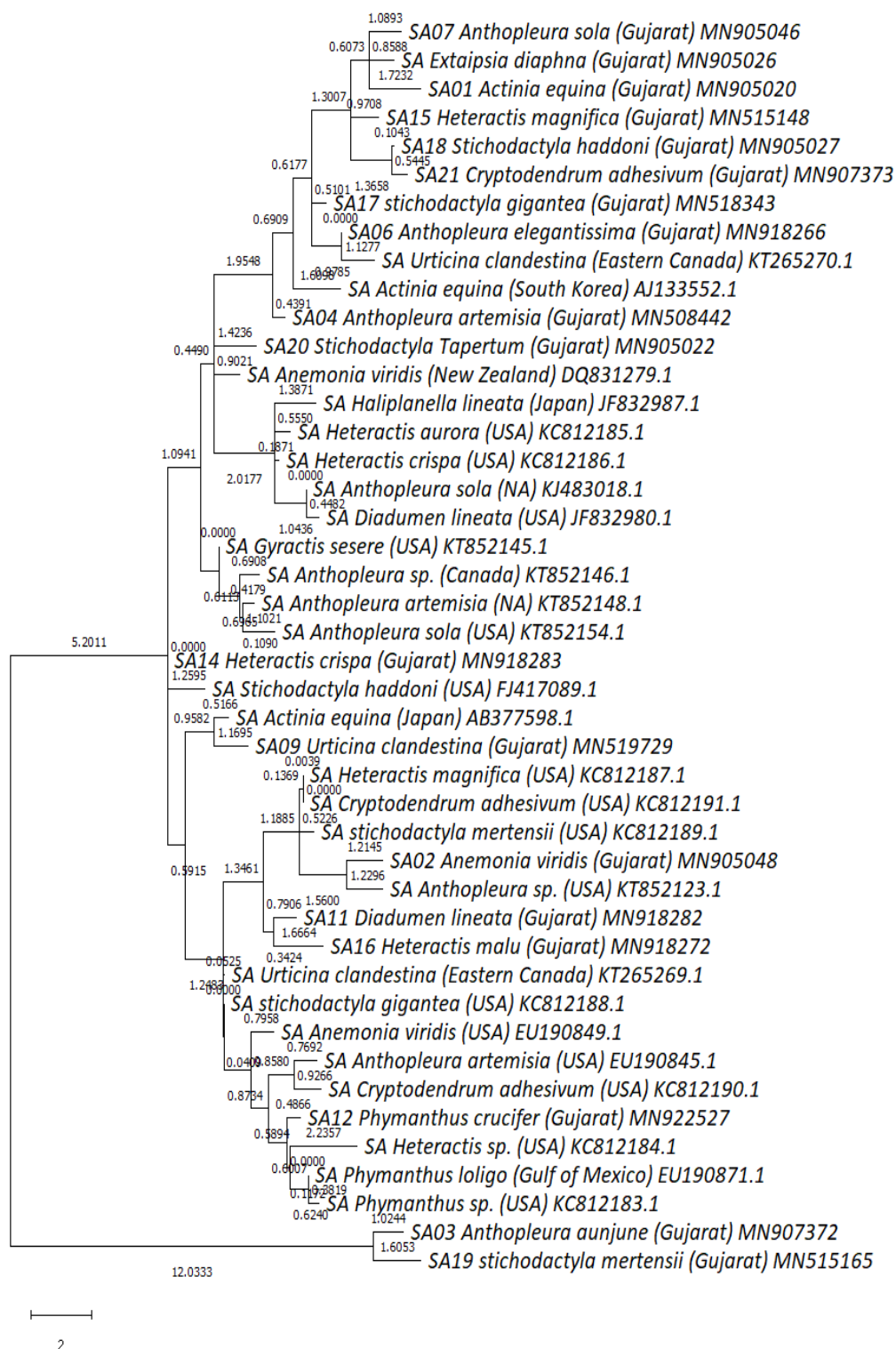


Figure 4. 4 : Phylogenetic tree for Sea anemone diversity comparing with worldwide fauna

Actinarians are conspicuous members of shallow-water environments. Although previously published phylogenetic studies of sea anemones

recovered only few of the family, they only included a very partial sampling in terms of its overall taxonomic diversity. However, no earlier study other than this has been documented for sea anemone molecular sequences from Gujarat. Therefore, the obtained sequence similarities were BLAST with the sea anemone species documented through the world and phylogenetic tree using Maximum likelihood method was prepared to analyse the relationship among them at species level (Fig. 4.3).

The evolutionary history was inferred using the Neighbor-Joining method [1]. The optimal tree with the sum of branch length = 37.68513074 is shown. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (950 replicates) are shown next to the branches [2]. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method and are in the units of the number of base substitutions per site. This analysis involved 18 nucleotide sequences. Codon positions included were 1st+2nd+3rd+Noncoding. All ambiguous positions were removed for each sequence pair (pairwise deletion option). There was a total of 1768 positions in the final dataset. Evolutionary analyses were conducted in MEGA X (Kumar et al., 2018). The BLAST results of the 18s nuclear region from most of the species did not match 100% with any described species except one but initial morphological groupings agreed with the genetic results. Thus, Phylogenetic tree using Maximum likelihood method also generated for the 18S rDNA sequence for 18 species clustered with similar sequence of the same genus documented worldwide (Fig. 4.4).

The evolutionary history was inferred by using the Maximum Likelihood method and Tamura-Nei model (Tamura and Nei, 1993). The tree with the highest log likelihood (-78404.35) is shown. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using the Maximum Composite Likelihood (MCL) approach, and then selecting the topology with

superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. This analysis involved **44 nucleotide** sequences. Codon positions included were 1st+2nd+3rd+Noncoding. There was a total of 1837 positions in the final dataset. Evolutionary analyses were conducted in MEGA X (Kumar *et al.*, 2018).

Sea anemones from the Aiptasiidae family are conspicuous members of shallow-water environments, with several species widely used as a model system for cnidarian-dinoflagellate symbiosis and coral bleaching studies. Most of the genera of this family proposed were recovered as monophyletic units, except for the genus *Aiptasia*, now divided into *Aiptasia* and *Exaiptasia*, a finding also supported by morphological diagnostic characters. Within *Exaiptasia*, the key focus of the taxonomic revision, the authors analysed specimens within species other than that correspond to most of the recorded species localities. *A. pulchella* and *A. couchii* (still included in *Aiptasia*) and could not consider morphological differences separating species currently described, thus synonymizing all species of *Exaiptasia* as a single widespread group; *Exaiptasia diaphna* (Grajales, 2014).

The morphological identification of four species belonging to Stichodactylidae family viz, *Stichodactyla gigantean* (Forsskal, 1775), *Stichodactyla tapetum* (Hemprich & Ehrenberg, 1834), *Stichodactyla mertensii* (brandt, 1835) and *Stichodactyla haddoni* (Saville-Kent, 1893) have always been confused because of their similar appearances and minor differences in morphological characters. Among that, *Stichodactyla mertensii*, *Stichodactyla haddoni* and *Stichodactyla gigantean* were distinguished through 18S ribosomal marker while *Stichodactyla tapetum* shows the higher branch length (19.94 01). More on, the 18S sequence for this species have not found recorded previously on NCBI but on the bases of some distinguish morphological character, we have reported as new record and also range extension. While comparing with worldwide documented species, *Stichodactyla tapetum* does not fall into the same node containing the *Stichodactyla* genus.

Based on column texture and tentacle length in the two species *Heteractis malu* and *Heteractis crispa* belong to the genus *Heteractis* can be distinguished in the field (Fautin and Allen, 1997). Both of these characteristics may be misleading since the former is open to subjective interpretation if both species are not present side by side in the wild, whereas the latter may not actually be useful because the tentacles may be contracted at the time of collection / observation. Through molecular identification both species were able to discriminate.

Family Actiniidae have the highest species divergence with total 7 species confirmed by molecular analysis. *Anthopleura elegantissima* and *Anthopleura aunjune* depicts as sister species. *Anthopleura aunjune* is reported for the first time from Gujarat coast and submitted as new species and thus not showing similarities with worldwide recorded data at species level but well clustered with *Anthopleura* genus. *Anthopleura elegantissima* shows similarities with *Gyactis sesere* documented from USA. However, such differences may be due to species having similar appearance but recorded as different species. Here in, the sequence documentation of *Anthopleura elegantissima* was done on the basis of evidence of its genus distribution.

Anthopleura artemisia (Pickering in Dana, 1848) is the first time reported from Alang site (Bhavnagar district) and presence of this species confirms the extended distribution of this species towards Indian Ocean. Moreover, presence of this species exhibits its capacity to survive in polluted conditions in Alang area.

Sea anemone species can be difficult to identify, especially in the field. Most of the taxonomic key used is based primarily on the presence / absence of specific characters or on historical differences and therefore requires the collection of whole animals, which may not always be practical. Moreover, histological analysis of marine anemones is time-consuming and requires considerable expertise as it is almost impossible for the non-specialist to identify certain closely related species, often resulting in an incorrect

taxonomic identification (Stephenson, 1928; Fautin, 2000; Hausser 2004; Wilmer and Mitchell, 2008).

Alternatively, the delineation of some species can be achieved quickly using suitable molecular genetic methods such as barcoding. Sea anemones contain very few studies examining the various barcoding regions (Dohna and Kochzius, 2016; Worthington and Mitchell 2008). The substitution rate in nuclear gene of Actinarian is much higher than in mitochondrial genes, having greater utility in terms of identifying species. Furthermore, a number of other studies have suggested that the complex of the nuclear ribosomal (rDNA) genes containing 18S, ITS1, 5.8S, ITS2 and 28S could be ideally suited for the study of the Actinarians at genus level relationships (McCommas 1991; Odorico and Miller 1997). In present study, we have examined the utility of 18S marker for species level identification of sea anemone. Nevertheless, surprisingly low divergence between two species of few genera, neither of which could be confirmed by histology due to the lack of availability of specimens, suggested that conventional histological methods are still needed to confirm identification. Finally, a number of aspects of this study reinforce the value of being able to combine histological analysis with genetic testing to irrefutably verify a species identification.

4.3 DISTRIBUTION OF SEA ANEMONES ALONG THE COAST OF SAURASHTRA

Sea anemones are the members of third large order Hexacorallia of Phylum Cnidaria and an integral part of coral reef ecosystem. The term sea anemone encompasses Order Actiniaria, a moderately diverse taxon. They inhabit marine and estuarine ecosystem, from upper littoral to abyssal depths, from burrowing to adhesive form and from free living to commensal types, thought accounting for a small part of the diverse marine biota of Gujarat.

This study encompasses the spatial distribution pattern of sea anemones at special level along the Saurashtra coast of Gujarat. Total 21 species of Sea

anemone belonging to 11 genera and 5 families are recorded from the Saurashtra coast of Gujarat. Family Actinidae contributes the 9 species belonging to 5 genera, Family Stichodactylidae contributes the 7 species belonging to 2 genera, Family Phymanthidae contributes the 2 species belonging to 1 genus, Family Diadumenidae and family Thalassianthidae contributes with single species each (Figure: 4.5). Herein, 13 out of 21 species of sea anemones are reported for the first time from Gujarat coast.

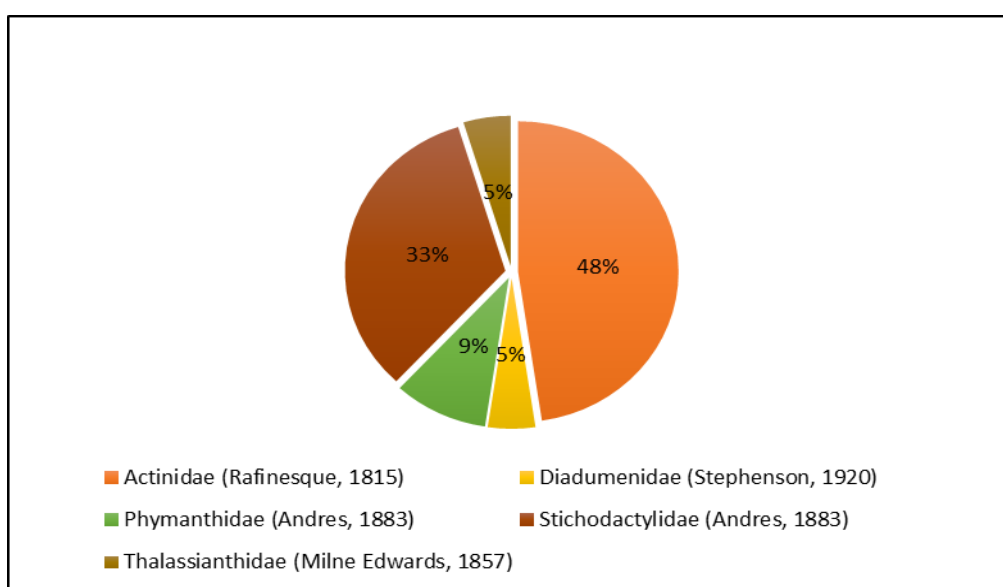


Figure 4.5: Contribution of Actinarian families recorded from Saurashtra coast

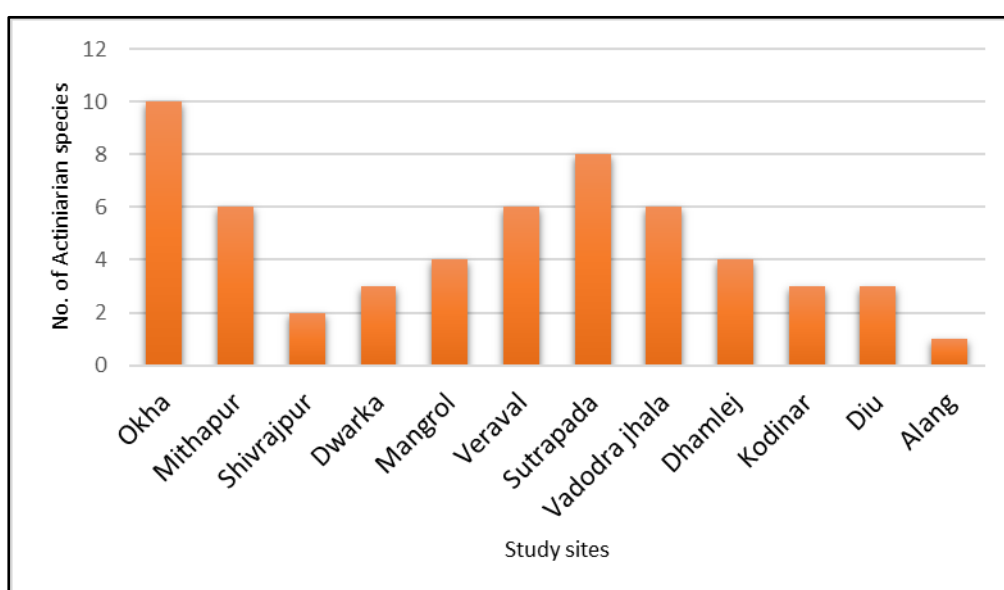


Figure 4.6: Site wise Actinarian species diversity

Total 12 sites have been surveyed from Okha to Bhavnagar. Among all the study sites, Okha coast represent the highest number of Actiniarian species having sandy habitat characteristics followed by Sutrapada with Rocky habitats (Figure: 4.6). Spatial distribution patterns of 21 sea anemone species have been studied using GIS technique using point scale maps (Fig.4.7-4.19).

Family Actinidae contributes the 9 species belonging to 5 genera. Rocky Intertidal zone of south Saurashtra coast supports the rich diversity of Actinidae family as these sites having large number of tide pools with very few sand patches. Among Actiniidae family, *Anthopleura* genus have higher number of species variation then others. *Anthopleura anjune* is the first-time recorded species from Gujarat. Previously, it was recorded from Goa by Hartog and Vennam, 1993 and reported as misidentification of *Anthopleura midori* (Uchida & Muramatsu, 1995). The sea anemone, *Anthopleura elegantissima* (Brand, 1835), was documented first time from the mid littoral zone of the coastal area of Vadodra jhala, on the coast of Saurashtra, Gujarat (Shah *et al.*, 2017). No records on this species and its aggregation from Indian waters are available to date.

Sea anemone, *Urticina cledenstina* belongs to Genus *Urticina* is commonly found in the upper and mid littoral zone at Vadodara-jhala of the Saurashtra coast is the new record of sea anemone fauna and range extension documentation (Shah *et al.*, 2015). Its presence in shallow tide pools in the intertidal zone makes them at risk to various environmental stresses during both low and high tide. This study also provides evidence of the adaptive strategy of *Urticina cledenstina* for the survival against the environmental stresses. The adherence of the gravel and shell to the external surface of the organism is the survival peculiarity for imitating surrounding habitat. This facilitates the camouflage. This species dominantly recorded from the Mangrol, Veraval, Sutrapada, Dhamlej and Kodinar. From North eastern Saurashtra coast, this species only found from the Dwarka coast due to the Clift structure of intertidal zone. *Actinia equine* belong to genus *Actinia*

distributed strictly to the southern Saurashtra coast. This species is mostly found in groups within tidepools or hanging on rock from upper and mid littoral zone. *Anthopleura sola* and *Anthopleura dixoniana* distributed through the south Saurashtra coast.

Anemonia viridis belong to genus *Anemonia* dominantly found from shivrajpur coast as rocky structure of upper zone supports the growth of this anemone hiding themselves in crevices and holes.

Here, for the first time, this study has reported the occurrence of species *Diadumene lineata* belong to Family Diadumenidae from the Saurashtra coast of Gujarat. Most likely as an anti-Lessepsian migrant from the Mediterranean Sea via the Suez Canal, considered to be one of the most spectacular biological invasions witnessed in the modern oceans. It is tough to imagine when the species came here; it could be a relatively recent arrival, most likely fouling in ship hulls. Occurrence of this species supports the possible migration in Saurashtra coast as Veraval is considers has one of the largest fishing center of India. However, this species has also been reported from Pakistani coast of Indian water (Kazmi, 2016).

Two Sea anemone species, *Phymanthus crucifer* and *Phymanthus buitendijiki* belong to Family Phymanthidae reported first time from sandy-rocky intertidal zone of Okha coast. Among this, distribution of *Phymanthus buitendijiki* was also reported from the South Andaman Island (Raghunathan *et al.*, 2014). This species can be easily identified on the bases of tentacle morphology. Tentacles are highly branched, brightly coloured and directly attached small flower like projections with tapered tips.

Family Stichodactylidae can be considered as second dominant and diverse family comprising of 7 species. The intertidal belt of okha and Mithapur greatly support the members of genera *Heteractis* and *Stichodactyla* as intertidal zone interspersed with various microhabitats such as shallow tide pools, crevices, coral pools zoanthid bed and algal bed. The morphological identification of four species, *Stichodactyla gigantean*, *Stichodactyla tapetum*, *Stichodactyla*

mertensii and *Stichodactyla haddoni* belonging to Stichodactylidae family have always been confused because of their similar appearances and minor differences in morphological characters. Till date these species were commonly reported as carpet anemones considering the species, *Stichodactyla haddoni*.

Hartog and Vennam, 1993 also reported the distribution of this species from Okha coast. On the bases of detailed study on species discriminating characters for these four species, we have reported them as different species with their taxonomical details.

Diversity and distribution of sea anemone species *Cryptodendrum adhesivum* belongs to the family Thalassianthidae reported first time from Okha coast. The appearance of this species is like Carpet anemones but can be distinguish on the bases of stickiness and morphology of anemones. Distribution record of this species documented thought the Andaman Nicobar Island reported by Raghunathan *et al.*, 2014.



Figure 4.7: Map showing point scale distribution of sea anemones at Okha coast

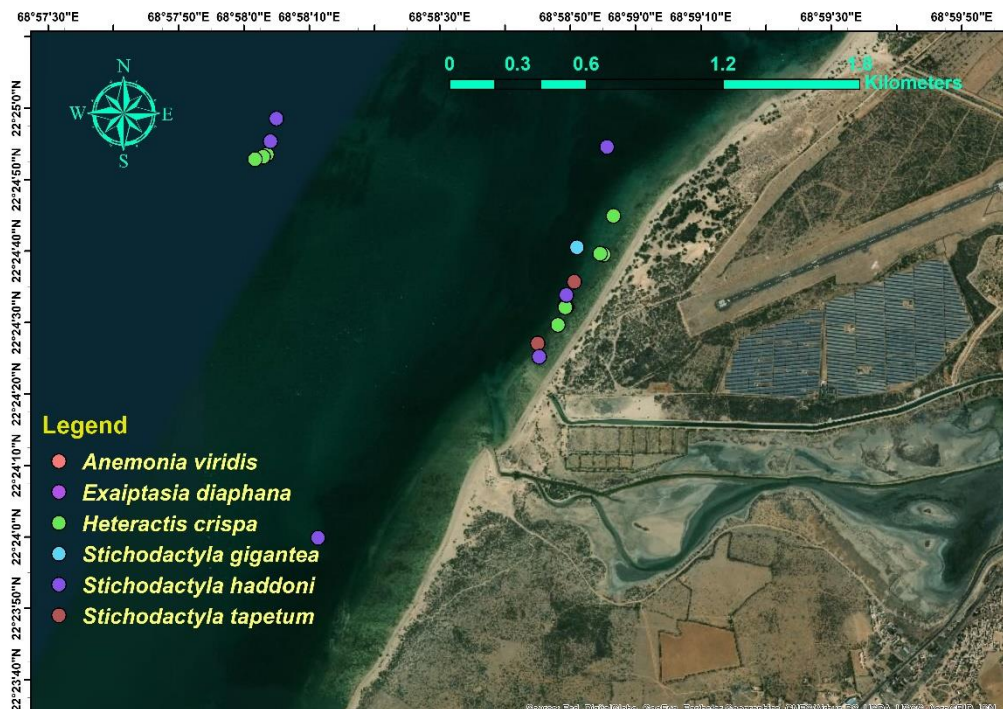


Figure 4.8: Map showing point scale distribution of sea anemones at Mithapur coast

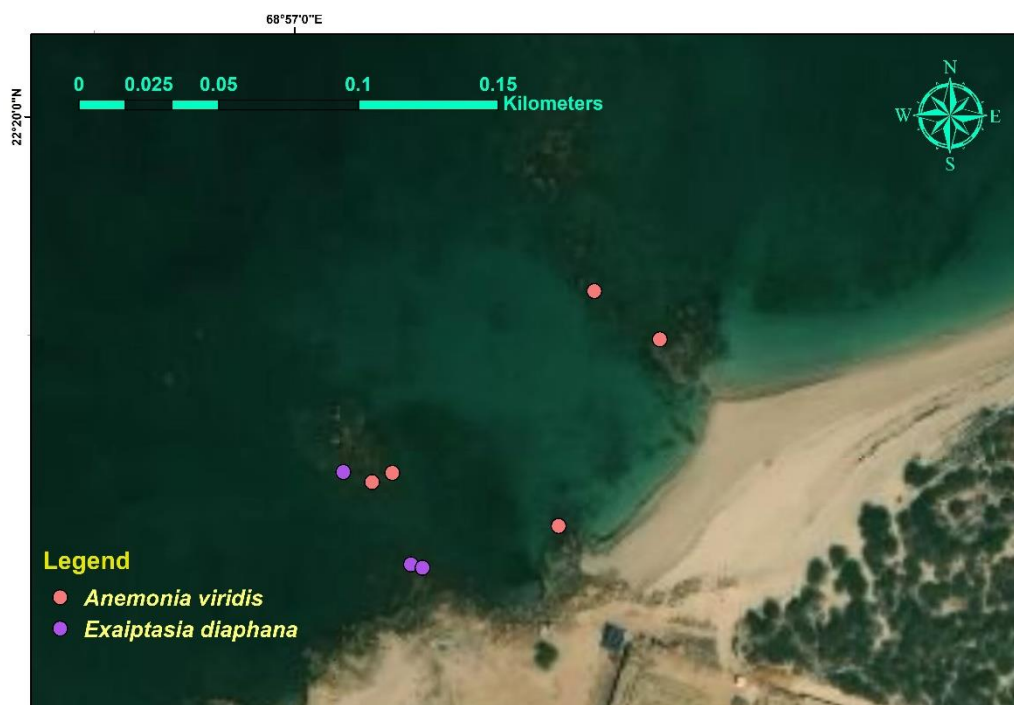


Figure 4.9: Map showing point scale distribution of sea anemones at Shivrajpur coast



Figure 4.10: Map showing point scale distribution of sea anemones at Dwarka coast



Figure 4.11: Map showing point scale distribution of sea anemones at Mangrol coast

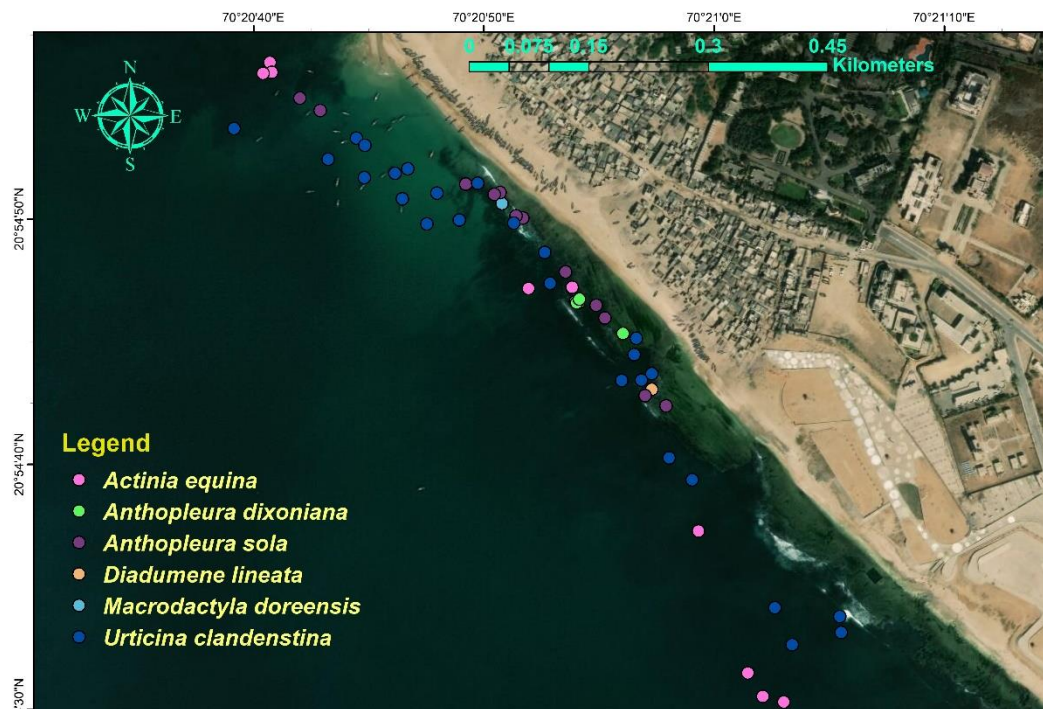


Figure 4.12: Map showing point scale distribution of sea anemones at Veraval coast



Figure 4.13: Map showing point scale distribution of sea anemones at Sutrapada coast

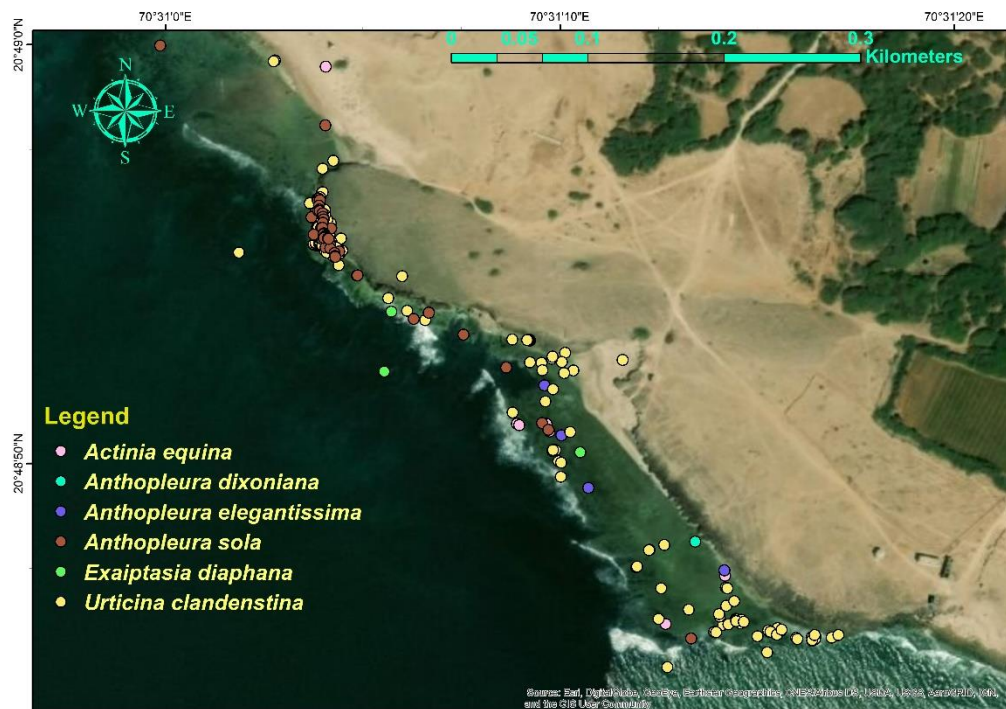


Figure 4.14: Map showing point scale distribution of sea anemones at Vadodra jhala coast



Figure 4.15: Map showing point scale distribution of sea anemones at Dhamlej coast



Figure 4.16: Map showing point scale distribution of sea anemones at Kodinar coast



Figure 4.17: Map showing point scale distribution of sea anemones at Jalndhar beach, Diu coast

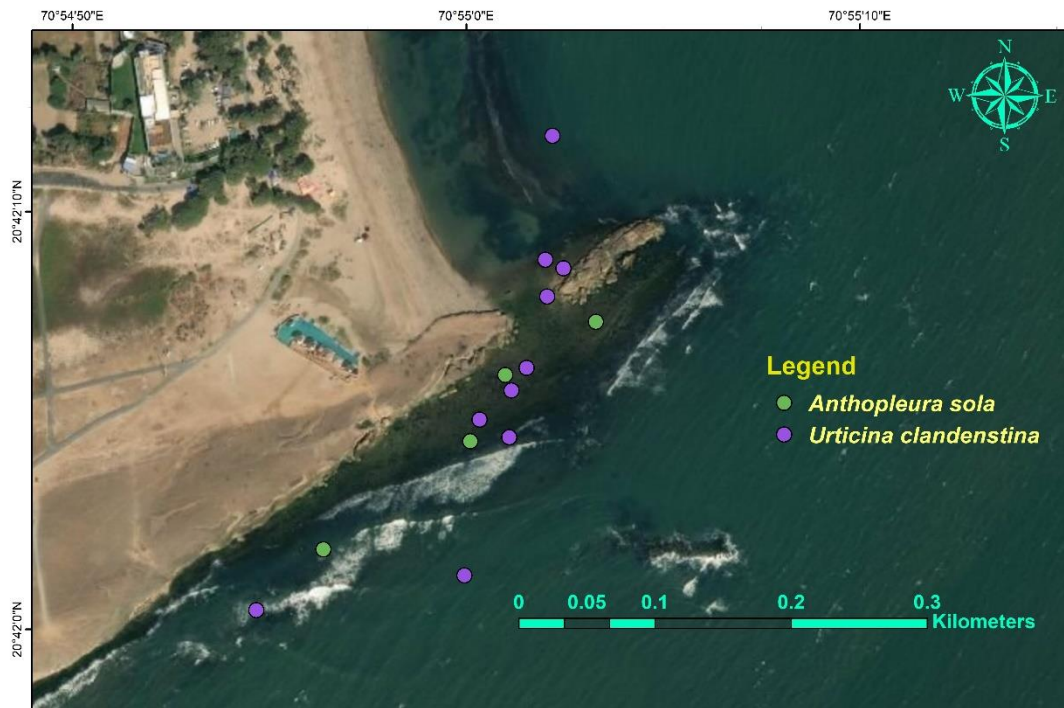


Figure 4.18: Map showing point scale distribution of sea anemones at Nagoa beach, Diu coast



Figure 4.19: Map showing point scale distribution of sea anemones at Alang coast

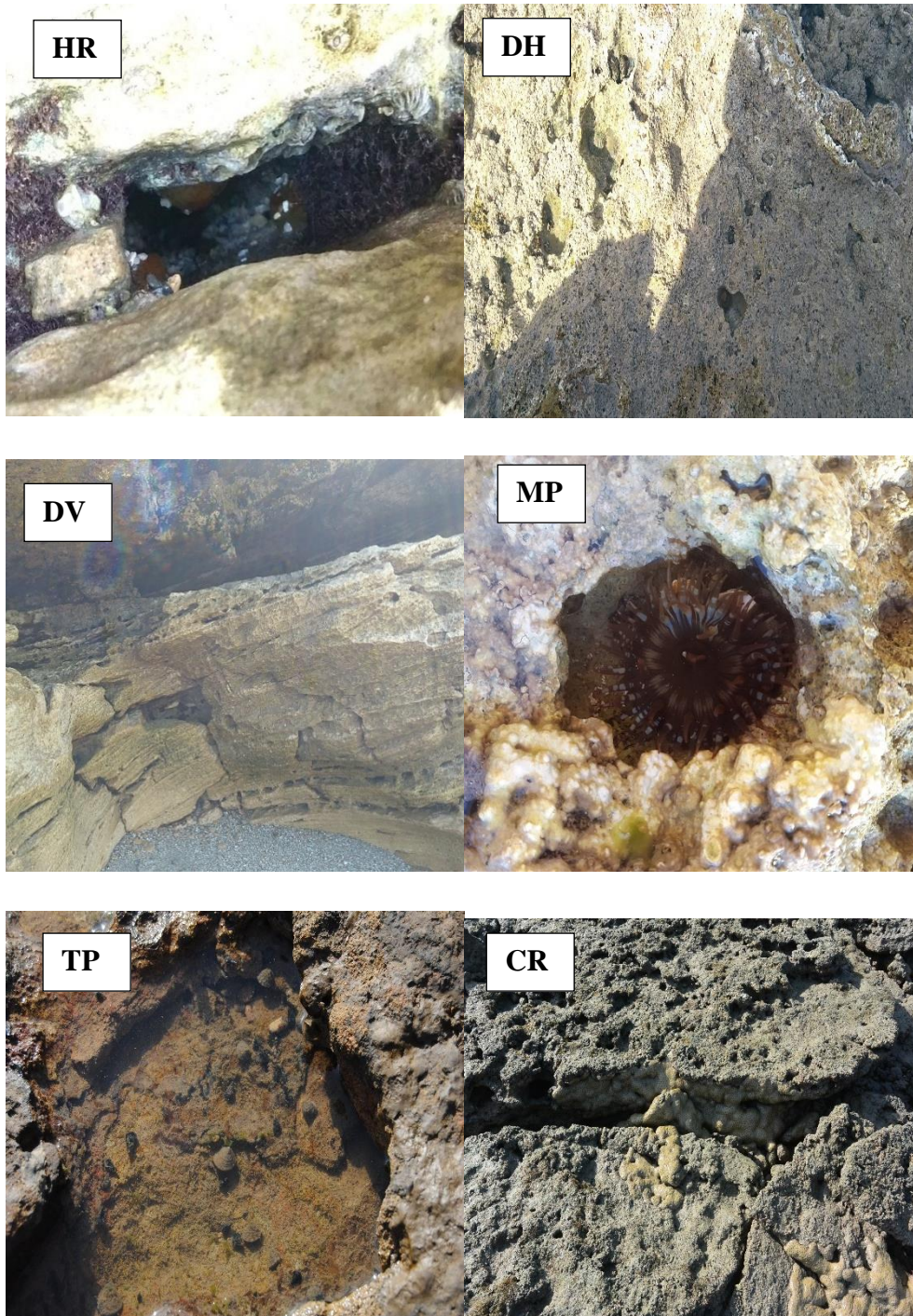
4.4 HABITAT CHARACTERIZATION AND PREFERENCES OF ACTINIARIANS

Intertidal rocky shores are particularly heterogeneous environments for different marine animals. The rocky intertidal group has been extensively studied for various ecological elements for two main reasons, such as simple exposure to the ecosystem and a rich natural resource. However, considering the long history of numerous ecological studies performed on rocky shores, many ecological issues concerning the spatial-temporal pattern of distribution of inhabited diversity communities are not described. Some organisms living in these ecosystems can be used as bio-indicators of environmental health or human activity, such as the community of sea anemones. Many researchers have found that the main trend of spatial aggregation of anemones is affected by light conditions, tidal oscillations and sea currents (Ottaway and Thomas, 1971; Pearse, 1974). Other related factors that may influence anemone attachment behaviour and patterns of distribution are still unknown, such as its habitat preference. For these reasons, our goal was to know the habitat preference of Actinarians so that future studies on anemone could track the presence of this bioindicator in coastal areas.

The Saurashtra coast is being hot spot for marine faunal and floral diversity. For this study, we were surveyed Saurashtra coastline and studied intertidal habitat characteristics. The substratum of this coast consists mainly of rocks of miliolite and laterite stones that provide a completely different ecosystem to sustain intertidal flora and fauna. The Saurashtra coastline is basically rocky, sandy, rocky-sandy, and muddy-sandy being rockier in the east and west, sandier in the central part and more rocky, muddy in the far eastern part. Thus, entire intertidal zone of the coast having different substratum, structures and abiotic factors.

Habitat assessment of sea anemone revealed very many differences in their habitat composition as all the selected study sites shows the selected types of microhabitats. Based on habitat classification scheme, the habitat preferences of sea anemone were divided into micro habitats for selected sites along the Saurashtra coast. The lack of difference among habitat was also apparent

when the shore surveyed for the detail habitat assessment were assessed using more detailed classification scheme 9 sub habitat at sea anemone preference; Hanging on rock under shelter (HR), dry hole at the base (DH), dry hole on vertical surface (DV), micro pool (MP), tidepool (TP), crevices (CR), sandy (SA), buried on sand but partially attached with rock (SR), muddy (MD) (Plate: 4.28).



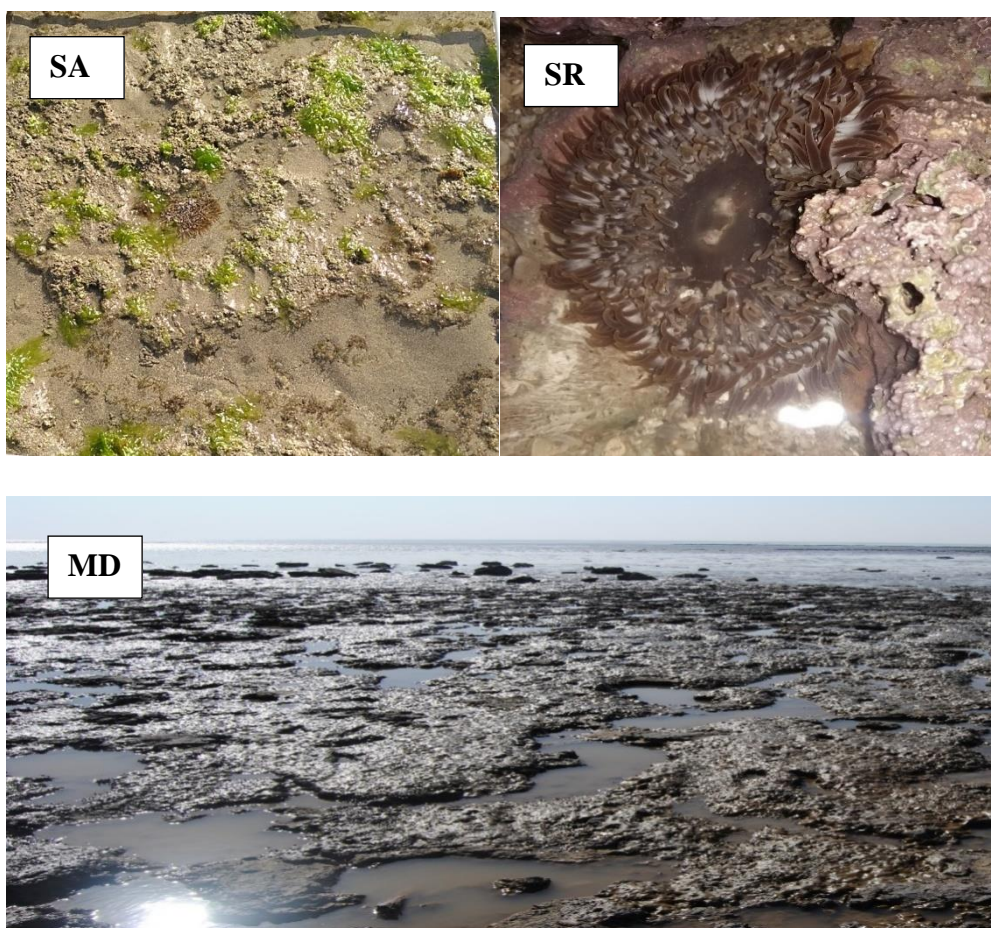


Plate 4.28: Different microhabitats preferred by sea anemone

Intertidal sea anemone diversity and habitat preferences attributes of 21 species belonging to 9 families and 11 genera were reported from the intertidal zone of Saurashtra coast. The microhabitat preferences of Actinarians were recorded on the bases of their occurrence at the different study sites (Table: 4.2).

| Sea anemone species | Different microhabitat preference | | | | | | | | |
|----------------------------|-----------------------------------|----|----|----|----|----|----|----|----|
| | HR | DS | DV | MP | TP | CR | SA | SR | MD |
| <i>Actinia equina</i> | + | - | - | + | + | + | - | - | - |
| <i>Anemonia viridis</i> | + | - | - | - | + | + | - | - | - |
| <i>Anthopleura anjunae</i> | - | - | - | + | + | - | - | - | - |

| | | | | | | | | | |
|----------------------------------|---|---|---|---|---|---|---|---|---|
| <i>Anthopleura artemisia</i> | - | - | - | - | - | + | + | - | + |
| <i>Anthopleura dixoniana</i> | - | - | - | + | + | + | - | - | - |
| <i>Anthopleura elegantissima</i> | - | + | - | + | + | - | - | - | - |
| <i>Anthopleura sola</i> | + | + | - | + | + | - | - | - | - |
| <i>Macrodictyla doreensis</i> | - | - | - | - | - | - | - | + | - |
| <i>Urticina clandenstina</i> | + | + | + | + | + | - | - | - | - |
| <i>Exaiptasia diaphana</i> | + | - | - | - | + | + | - | - | - |
| <i>Diadumene lineata</i> | - | - | - | + | + | - | - | - | - |
| <i>Phymanthus crucifer</i> | - | - | - | - | - | - | + | - | - |
| <i>Phymanthus buitendijki</i> | - | - | - | - | - | - | + | - | - |
| <i>Heteractis crispa</i> | - | - | - | - | - | - | + | + | - |
| <i>Heteractis magnifica</i> | - | - | - | - | - | - | - | + | - |
| <i>Heteractis malu</i> | - | - | - | - | - | - | + | + | - |
| <i>Stichodactyla gigantea</i> | - | - | - | - | - | - | + | + | - |
| <i>Stichodactyla haddoni</i> | - | - | - | - | - | + | + | + | - |

| | | | | | | | | | |
|---------------------------------|---|---|---|---|---|---|---|---|---|
| <i>Stichodactyla mertensii</i> | - | - | - | - | - | + | + | + | - |
| <i>Stichodactyla tapetum</i> | - | - | - | - | - | + | + | + | - |
| <i>Cryptodendrum adhaesivum</i> | + | - | - | - | - | + | - | - | - |

Table 4.2: Different micro habitat preferences of sea anemone based on their occurrence

Bray-Curtis similarity percentage was calculated to find out the similarity within species diversity between different types of microhabitat. Results show that the similarity in species composition between different microhabitats ranges from 74% to 0%. Here mainly three groups formed by different microhabitats. First group is formed by microhabitats like MP, HR, TP, DH and DV. Second group is formed by microhabitats like CR, SA, and SR. The third group is formed by Muddy habitat, MD joining rest of the groups.

Maximum similarity (74%) in terms of species composition was observed for microhabitats like DH and DV forming a separate group. MP and HR show 53% similarity in species composition. TP microhabitat joining with 28% similarity to group formed by MP and HR strongly represent the rocky habitat. Second group is formed by SA and SR with 55% similarity. Microhabitat CR joining this group with 38% similarity. Micro habitat like MD formed a third group with 0% similarity with another microhabitat because only *Anthopleura artemisia* utilizes this microhabitat and sharing the microhabitat like TP and CR.

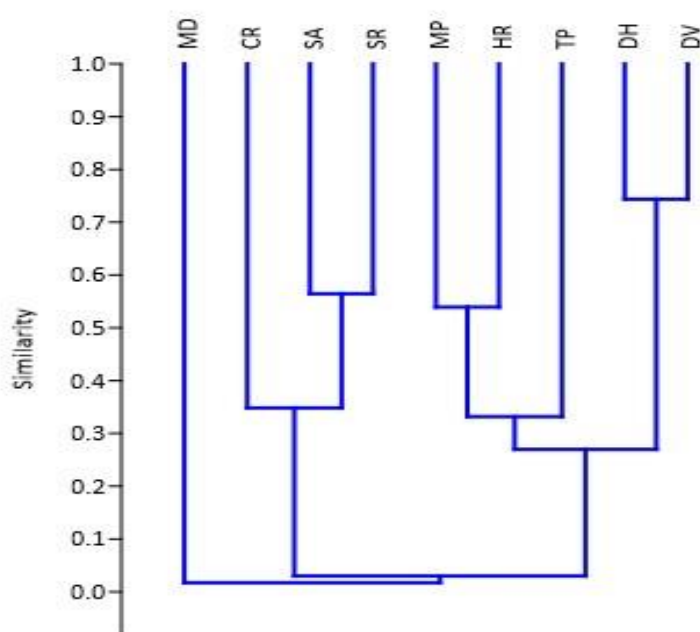


Figure 4.20: Bray- Curtis similarity indices for Actinarian diversity of different microhabitats

Among the sea anemone diversity, members of Family Actiniidae show the highest occurrence in various habitat structure followed by Stichodactylidae family (Fig. 4.21). Genus *Actinia*, *Anthopleura* and *Mycodacyla* belongs to Family Actinidae strongly preferred the rocky substratum of Southern Saurashtra coast habituated in micro habitat such as hanging on rock under shelter, dry hole at the base micro pool, tidepool and crevices. *Anthopleura Artemisia* belong to family Actinidae mostly found habituated in sandy muddy habitat of eastern part of coast. Sediment depositions increase towards the lower intertidal zone: having least deposition in upper intertidal zone (Fig. 4.22). Sediment deposition in lower intertidal zone is so large, that makes it appear muddy. It has been documented earlier that *A. artemisia* never has symbiotic algae in its endoderm (Shick *et. al*, 2002). This can be a possible reason for abundance of non-symbiotic *A. artemisia* on the Alang site.

Urticina clandenstina belongs to family Actinidae founds only species of this family, which is distributed all over the coastal area of Saurashtra. However, this species mostly preferred the microhabitat such as hanging on rock under shelter, dry hole at the base, micro pool, tidepool and crevices of South coast; while most dominantly preferred the micro pool, tidepool and on dry hole of vertical surface situated at Dwarka rocky coast having wide and deep caves on outer intertidal zone, highly elevated due to heavy wave action.

Anemonia viridis belong to family Actinidae has shown preferences to the north-eastern coast with rocky sandy substratum. This species mostly found in the micro habitat such as hanging on rock under shelter, tide pool and crevices. *A. viridia* also seems to prefer pools with a medium and high number of caves, and these caves or fissures could provide a more protective environment and a better ecological condition. It looks like *Anemonia sp.* preferred habitats are tidal pools with caves where they grow carnivorously in balance with their photosynthetic activity of zooxanthellae having yellow colouration of tentacles. When these anemones live well outside caves, however, they are vulnerable and have a higher incidence of the light, thus showing different colours (Taylor, 1967, 1969; Gonzalez-Delgado et al., 2018). This is because they have more symbiotic zooxanthellae that give a darker colour to them. Such studies indicate the positive relationship of colouration of species with respect to micro habitat preferred by them. *Diadumene lineata* belong to Family Diadumenidae has reported low occurrence from the rocky shore of Sutrapada and Veraval habituated in micro pool and tide pool. In literature, it is frequently found in places near harbours, pilings, and floats or shipping lanes. Also occurs in brackish-water creeks and lagoons, in estuaries, etc. (Molina *et al.*, 2008). To minimize exposure and possible desiccation, it can retract its tentacles and contract into globular hemisphere in mucus and this trait may enhance the ability of the species to survive long distance accidental transport (Cohen, 2005).

Family Stichodactylidae found to be predominant on Okha and Mithapur, having rocky with sand deposited topography. The members of genus

Heteractis and *Stichodactyla* mostly found lived attached firmly to the substrate by its pedal disk with the column buried in sandy substrate. Sea anemone species *Phymanthus crucifer* and *Phymanthus buitendijki* belong to Family *Phymanthidae* mostly found column buried in sand at the sandy shore of Okha.

Exaiptasia diaphana belongs to family *Aiptasiidae* distributed throughout the northern, eastern and south Saurashtra coast. This species preferred the micro habitat such as hanging on rock, tide pool and crevices of shore. The high tide line and the supra littoral fringe are on a sloping sand beach, but the upper intertidal zone of okha is formed by a pitted conglomerate lime-stone outcrop (Kohn, 1969). *E. diaphana* habituates such upper intertidal zone found highest within crevices of this coast.

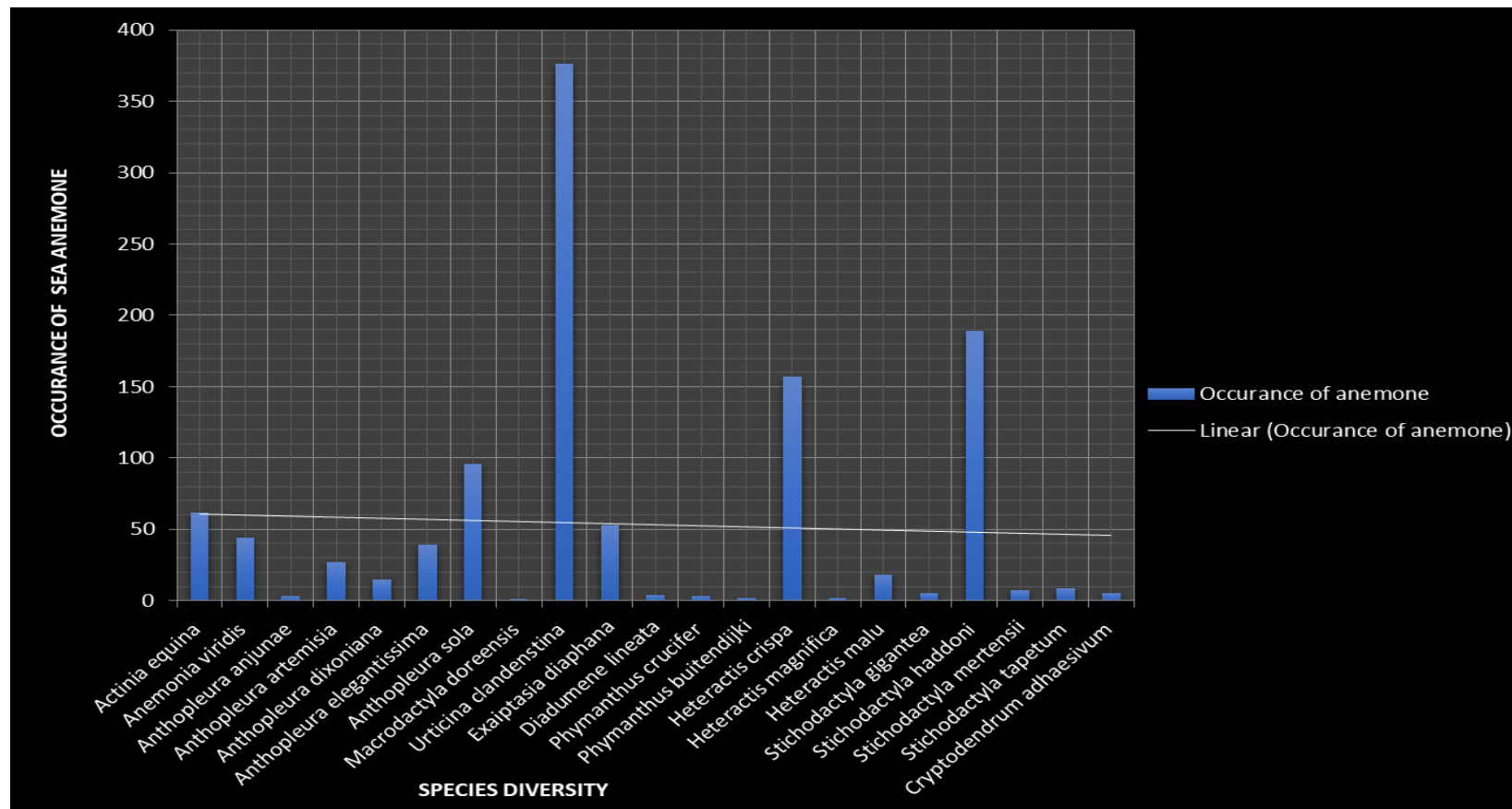


Figure 4.21: Diversity of Sea anemone occurrence along the Saurashtra coast

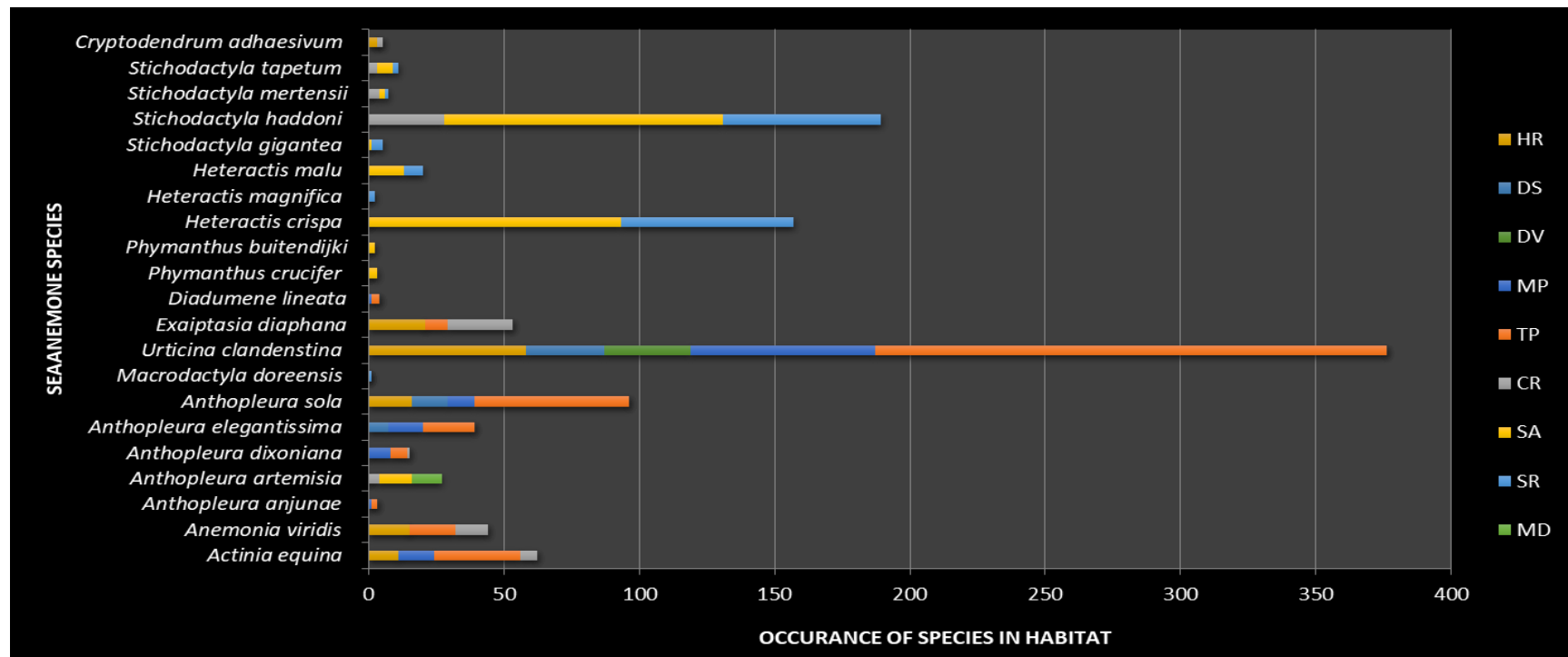


Figure 4.22: Sea anemone microhabitat preferences along the coast of Saurashtra

Hanging on rock under shelter (HR), dry hole at the base (DH), dry hole on vertical surface (DV), micro pool (MP), tidepool (TP), crevices (CR), sandy (SA), buried on sand but partially attached with rock (SR), muddy (MD)

Marine intertidal species often disperse large distances of planktonic larvae. Therefore, larvae need to have mechanisms for locating recognizing, and settling in habitat types suitable for juvenile development, survival, and reproductive success. Specific invertebrate larvae substratum selection has been shown for species in a variety of phyla (Knight-Jones, 1953) but less attention has been paid to the secondary habitat selection process by settled individuals. Completely sessile species cannot change their location if it proves to be unsuitable, so the habitat chosen by a larva is also the adult habitat. However, more mobile animals' juvenile habitat may be different from that of adults (Gale, 1971). Sea anemones can move several centimetres a day (Parker, 1919) and thus have the option of changing location throughout their lifetime. Such slow movement showing sea anemones mostly found in rocky and sandy habitat, lives under the rock crevices and tide pools.

Intertidal environments are biologically and physically harsh, with stressful environment frequently enhanced by organisms forming the resident ecosystem (Menge and Sutherland, 1987). Sea anemone fauna and habitat preferences have always ignored despite being often abundant macro faunal communities that live in the biogenic habitat created by the habitat forming organisms. The improvement of stressful conditions associated with desiccation and wave stress, with strong relationships between habitat structure and complexity, and the diversity and abundance of this fauna, could be expected to be important for these macro faunal organisms. Such positive relationships between biodiversity and habitat structure are fundamental to many main ecological issues (e.g., facilitative interactions, niche aspect characterization) and have major implications in habitat mapping and conservation.

In the present work, it was observed that the availability of various micro habitat has immense impact on the Actinarian diversity of the coastal area. Micro habitat characteristics inhabited by each species are different. when it comes to specific species, we have observed its attachment with substratum and surrounding organisms. However, critical ecological comparison of

associates was not considered for analysis. Further studies are needed to find out the variation in the abiotic and biotic properties of specific micro habitat on the species in habiting that micro habitat.

4.5 SEA SURFACE TEMPERATURE (SST) AND SEA ANEMONE BLEACHING

Coral reefs are among the most vulnerable to climate change in all coastal ecosystems. A variety of environmental stress is known to cause worldwide coral bleaching, such as elevated sea surface temperature (SST), increased or decreased solar irradiation, reduced salinity and sedimentation (Hoegh-Guldberg, 1999). The impact of increased sea surface temperature was clearly evident at the North-eastern Saurashtra coast in the form of coral and sea anemone bleaching.

Occurrence of coral and sea anemone bleaching evident at both the sites, Okha and Mithapur. During the sea anemone diversity survey work from Saurashtra coast, four bleached sea anemone species were recorded during summer, 2016. Four species belong to two genera *Heteractis* and *stichodactyla* were found affected by thermal stress. The degree of bleaching differed between different colonies and even the same species showed a difference in intensity of bleaching. Most of the bleaching found in family Stichodactylidae: *Stichodactyla haddoni* and *heteractis crispa* found reported bleached from Mithapur; *Heteractis malu* and *heteractis crispa* from Okha (Plate: 4.29).

SST is a critical factor in the well-being of host animals such as corals and sea anemones. Determination of how SST affects the coral reefs is critical. SST anomalies demonstrate possible perturbation to coral reef ecosystems and sometimes expose potential changes in resources and abundance of organisms that could be prioritized for marine conservation efforts. The aim of this study was to use satellite data to monitor daily SST anomalies that trigger coral bleaching in India's Saurashtra coast region during summer 2016. For the present study, the SST data analysis was performed using two types of SST data products with different spatial and temporal resolutions.

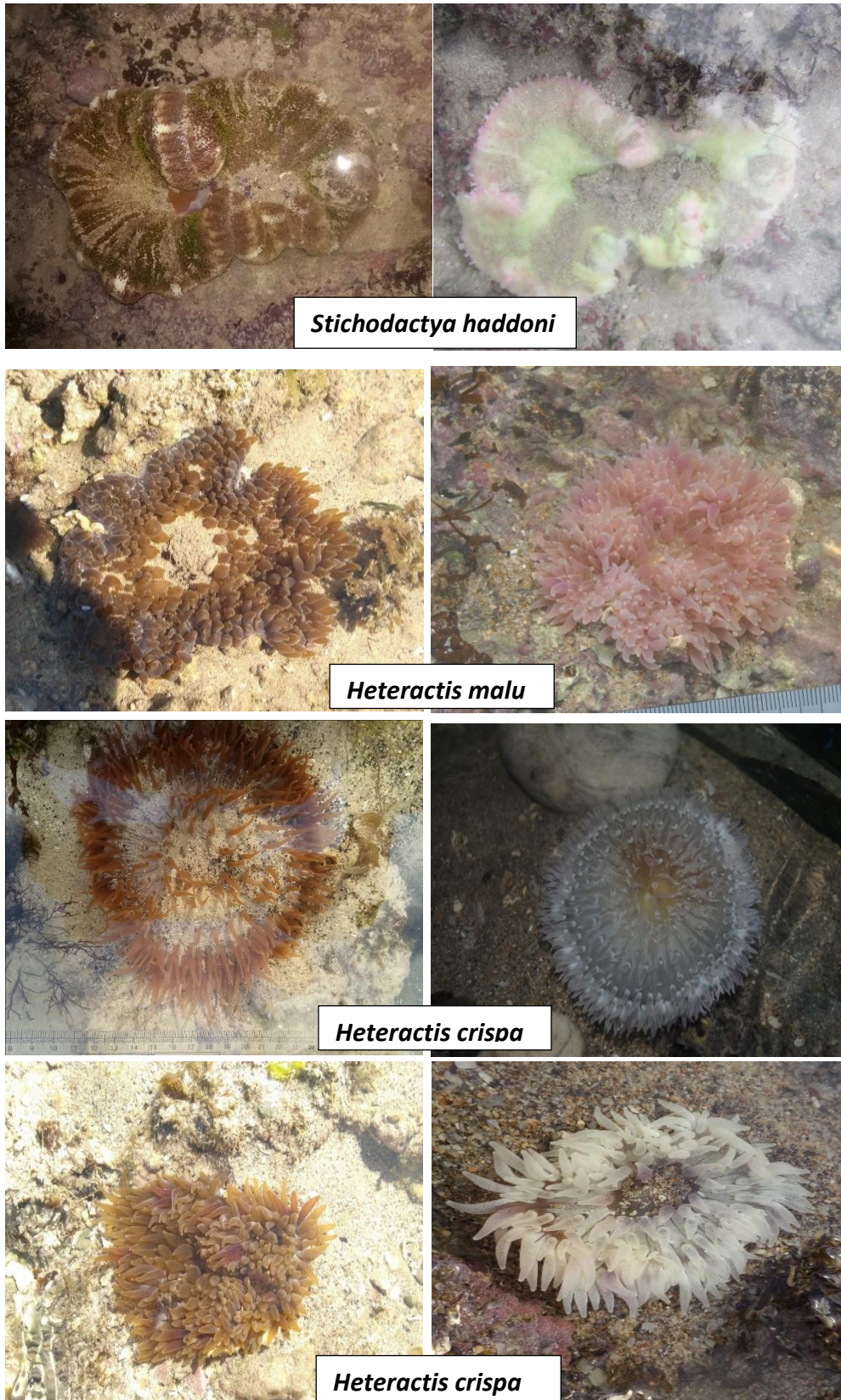


Plate 4.29: Bleaching found in sea anemones: Original condition (Left) and bleached (Right)

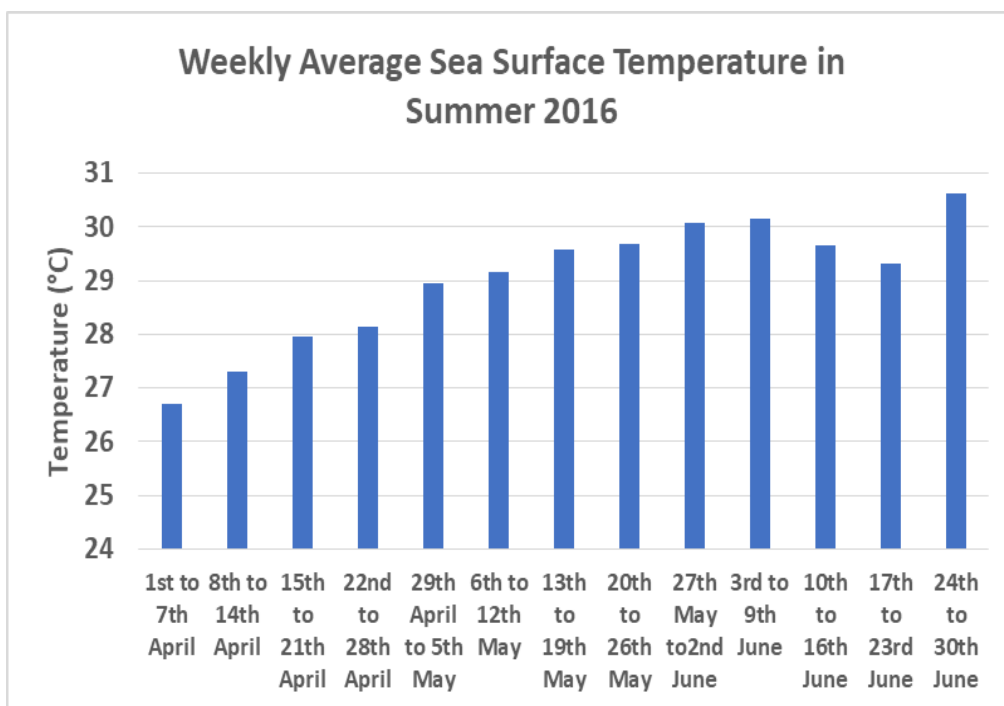


Figure 4.23: Weekly SST experienced for summer 2016 in Northern Saurashtra coast

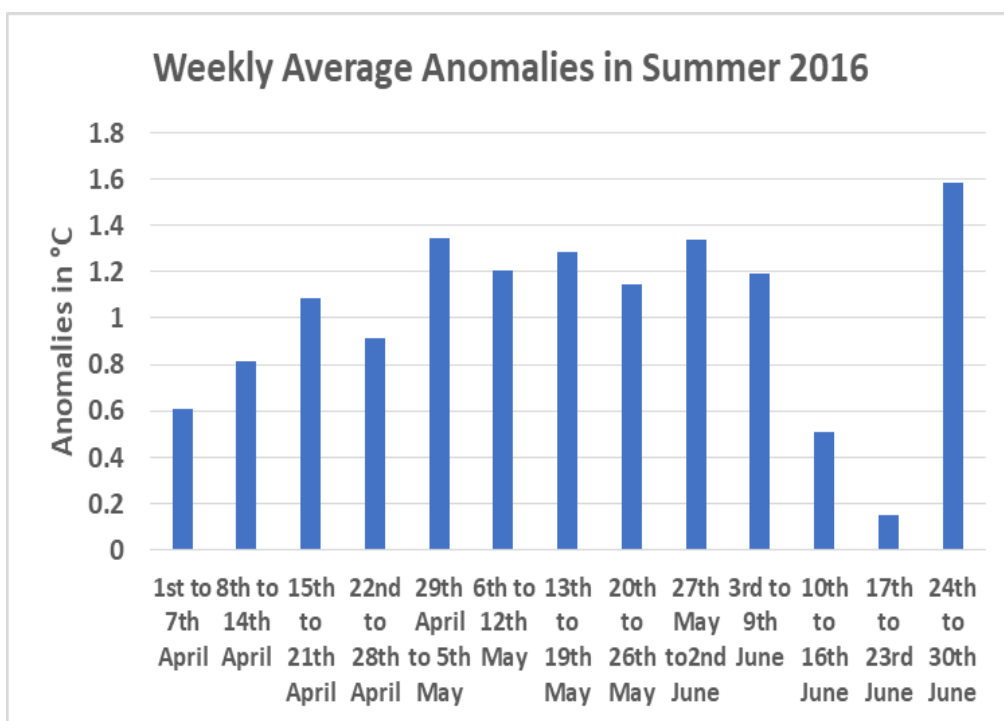


Figure 4.24: Weekly SST anomaly during April-June 2016 in Northern Saurashtra coast

This study demonstrated the warmest quarter of the diurnal trend of SST and daily SST anomalies derived from NOAA AVHRR data for 1st April to 30th June 2016 for the northernmost part of Saurashtra coast covering Okha and Mithapur coast.

In April 2016 the average value of daily SST recorded was 27°C. Which is well below the regional thermal threshold and could be considered as cooler week compared to the entire summer, 2016 (Fig. 4.23). The SST values recorded 29 °C, exceeded the thermal threshold during the first week of May and continued for two weeks. The SST rose further in the second half of May, and even crossed the threshold. Maximum SST of 30 °C was recorded in the month of June. In the second half of the June, SST came down. The SST condition became worsened during the last week of June, 23rd to 30th June. Thus, last week of June was also the warmest week of summer, 2016.

The temperature anomaly indicates a deviation from a reference value or a long-term average. Herein, Weekly average SST anomalies were recorded from 1st April to 30th June for Northern Saurashtra coast (Fig. 4.24). The first spell of anomalies recorded was 0.61 °C during the first week of April 2016. However, these anomalies tend to gradually increase in coming weeks of April, i.e. 0.8 °C and 1.1 °C during second and third week respectively. During the first week of May, highest anomalies of 1.6 °C was recorded, which started to be decreased in coming weeks. Last week of May and first week of June (27th May – 2nd June), anomalies tend to increase by 1.33°C. Again, in the mid of June, anomalies decreased by 0.15°C, which was the lowest value of entire summer, 2016. However, highest anomalies of 1.58 °C recorded during 24th to 30th June which is the warmest week of summer, 2016.

Sea anemone bleaching was reported during the first week of May from Mithapur and last week of June from Okha while surveying the sites for sea anemone diversity. Bleaching of *Stichodactyla haddoni* and *heteractis crispa* was recorded on 3rd May during the visit to Mithapur. On computing this field data with SST and SST anomalies, supports the possible impact of increased temperature on sea anemone in the form of bleaching. Although, SST during

the period of 29th April to 5th May was around 29°C, but according to higher SST anomalies of 1.34°C indicating 2nd highest anomalies week of summer, 2016. More on, from the Okha coast *Heteractis malu* and *Heteractis crispa* reported bleached on 28th June during field visit. However, average weekly SST data was recorded highest 30.63 °C during the 24th June to 30th June along with highest increase in anomalies which was reported 1.58 °C. Thus, SST data summarised that summer 2016 was warm enough to cause bleaching particularly in hard corals as well as in zoanthallate sea anemones.

Okha lies on the outer rim of the Kachchh Gulf and forms the northernmost portion of the Saurashtra coastline. As the Mithapur Bay is an integral part of the Gulf, their dynamics and ecology are influenced by the processes in the Gulf. Mithapur tides are mixed semi-diurnal with two unequal high waters and two unequal low tide water which occur every day. The tidal water flows into the Gulf from the south-west and affects Mithapur's north-west shore. The Gulf of Kachchh situated at the northern boundary of Saurashtra Peninsula of Gujarat represents the northern most limit of coral reefs in India. The reef is characterized by diverse coral species, which are potential bio-markers of the environmental stresses which gets reflected in the form of coral bleaching evidences. During the summer months of 2010, 2013 and 2014, the mass coral bleaching was observed to varying degrees on the GoK reefs. Joshi (2016) reported the 19 bleached coral species belonging to 13 genera and 7 families were affected in the GOK. The impacts of thermal stress were clearly evident in the form of mass coral bleaching at the reefs of the GoK. Furthermore, Arora et al., 2019 studied the daily SST anomalies triggering coral bleaching in the Gulf of Kachchh region of India during summer 2016. It was found that in summer 2016, SST rose to 30.62 °C and recorded a maximum positive anomaly of 1.31°C in the month of June supporting the result of this study from Northern Saurashtra coast. Coral bleaching was also reported attributable to thermal stress in the Andaman region during April 2016 (Mohanty et al., 2016). Bleaching was recorded from many coral reef sites around the world, including The Great Barrier Reef (GBR) which experienced the worst mass coral

bleaching event in its history in 2016. The GBR recorded its hottest-ever average SST for February, March, April, May and June since the historical 1900 SST records in 2016, according to the Bureau of Meteorology (Australia). Current warming anomalies show that the global extent of coral bleaching during 2015-2016 was a good "global bleaching event for coral reefs".

Synergistic use of geophysical parameters such as SST obtained from remote sensing satellite along with early field / *in-situ* monitoring of bleaching responses can aid in the timely prediction / pre warning of bleaching process. Monitoring of daily SST anomalies supported by real-time field monitoring of bleaching responses can help develop and adopt appropriate management strategies to alleviate the environmental stress on coral reef ecosystems.

4.6 THERMAL STRESS AND HSP (HEAT SHOCK PROTEIN) EXPRESSION

The environmental stresses associated with climate change are widely recognized as the most serious threat to the health of coral reefs (Hoegh-Guldberg *et al.*, 2007; Grottoli *et al.*, 2014). Over recent decades, climate shifts have intensified thermal stresses on corals, that have led to a worldwide decline in coral cover and dramatic changes in coral community structure (Hughes *et al.*, 2007; Hoegh-Guldberg and Bruno, 2010; Montano *et al.*, 2010; Death *et al.*, 2012). Sessile lower invertebrates such as sea anemones are directly exposed to extreme environmental conditions in certain marine habitats, such as elevated temperatures, which can cause tissue damage (Sharp *et al.*, 1994). In fact, although coral bleaching, namely the loss of endosymbionts and/or their photosynthetic pigments, occurs in response to several abiotic stressors (reviewed in Lesser, 2011), the main coral bleaching episodes that have resulted in significant mortality are strongly correlated with elevated sea temperatures (Smith *et al.*, 2008; Van Woesik *et al.*, 2011; Alemu and Clement, 2014). Stress proteins are expected to play an important role in giving those organisms tolerance to such harsh conditions. The response of

stress proteins, present in almost all the species studied so far involves the rapid synthesis of a group of proteins called Heat shock proteins (HSPs). In this context, understanding how temperature changes can affect the coral cellular mechanism involved in the stress response appears critically important. A ubiquitous biological process that consistently emerges as a putative marker of temperature-induced cell stress is the involvement of HSP, (Olsen *et al.*, 2013). HSPs exist in high concentration in the cell and fulfil a crucial role as a defence mechanism by sustaining protein homeostasis during cellular stress through the up-regulation of their expression (Kastle and Grune, 2012; Bozaykut *et al.*, 2014). In fact, HSPs facilitate proper protein folding and multimeric protein assembly, assist protein translocation, prevent the aggregation of misfolded proteins and facilitate the removal of protein aggregates, participate in signalling pathways and regulate stress-induced apoptosis (Hartl *et al.*, 2011; Chow *et al.*, 2012; Kim *et al.*, 2014; Seveso *et al.*, 2014).

In certain marine environments, lower sessile invertebrates such as sea anemones are directly exposed to harsh environmental conditions such as high temperatures, which may cause tissue damage (Sharp *et al.*, 1994). Stress proteins are supposed to play a significant role in conferring tolerance to such harsh environments on these species. The Saurashtra coast has stressful ecosystems, such as tidal pools. These are very restricted shallow water bodies that often become directly exposed to air without experiencing any change of water for several hours or days. Consequently, severe and unexpected changes in environmental conditions, particularly temperature, can occur. The coastal zone also features rocky environments, marked by temperate changes in the climate. Due to the exploitation of different ecological zones of rocky shores (tide pools and subtidal zones) by spatially distinct populations, sessile Cnidaria, such as sea anemones, provides an ideal group of organisms to examine adaptive responses to changes in environmental conditions.

Among the HSPs, we focused on the expression of HSP60 and HSP70 under different temperature conditions to gain insight into the response to thermal

stress in sea anemone at the molecular level due to the importance of these genes in the stress response. HSP60 and HSP70 are generally considered to contribute to stress responses (Nakamura *et al.*, 2012; Seveso *et al.*, 2014; Seveso *et al.*, 2016). HSP60 also have an important role in regulating the balance of mitochondrial-associated metabolic pathways in relation with environmental conditions (Papp *et al.*, 2003; Kingsley *et al.*, 2003). Therefore, evaluating the expression of HSP60 and HSP70 is a good starting point for investigating the relevancy of HSPs expression in sea anemones under thermal stress conditions with quantitative analyses.

Urticina clandenstina is the most abundant species found along the Saurashtra coast of Gujarat. This species is mainly found in the upper and middle intertidal region, consisting of many tidal pools that anatomize each other and form a wide network of aquatic regimes. This is the first study to quantify the heat shock response in *Urticina clandenstina*. In this study, we examined the ability of the sea anemone *Urticina clandenstina*, to express a HSP60 and HSP70 as a phenotypic adaptation (acclimatization) to changes in seawater temperature from the same tide pool where temperature layers appear. This protein is known to play a significant role in resistance to adverse temperatures (Cheng *et al.*, 1989). The temperatures always lower in the deep level of the sea. However, this will affect the HSP60 and HSP70 gene expression.

Urticina clandenstina, colonizing tidal pools that occasionally experience the generation of temperature layers separated by a thermocline. This situation arises when tidal pools are isolated from the open sea for 24 hours to several days. When temperature layers appear, different specimens from the same tidal pool are simultaneously exposed to different temperatures. *Urticina clandenstina* were observed along a vertical gradient. The sea anemone lives near the surface, and experience seasonal heating due to solar radiation. During such events, we collected samples from upper and lower parts of the tide-pool of Vadodra jhala study sites when the Sea Water Temperature (SWT) varies between 20°C-31°C and examined their levels of HSP60 and HSP70 expression.

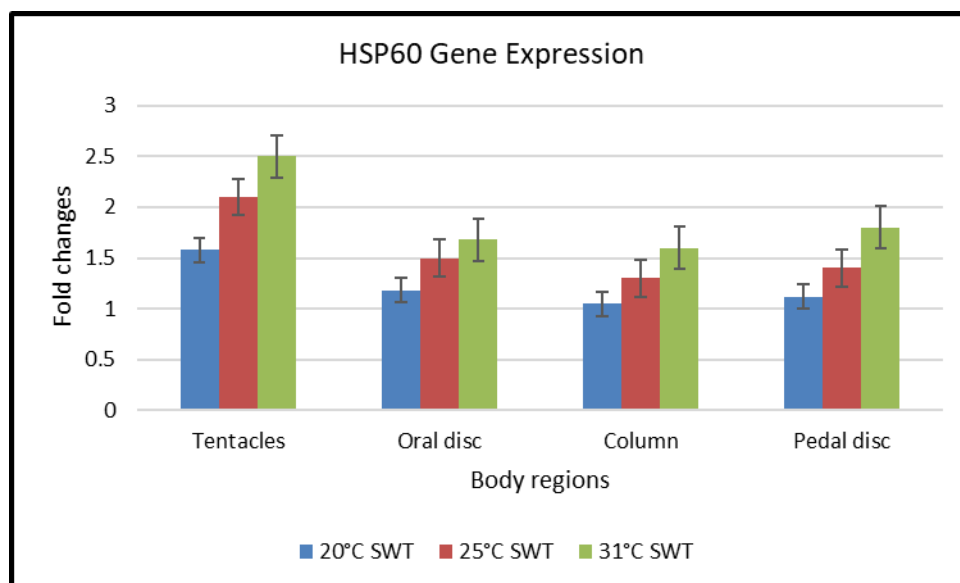


Figure 4.25: Variation of HSP60 gene expression in *U. claudenstina* body region (tentacle crown, oral disc, column and pedal disc) with increasing temperature

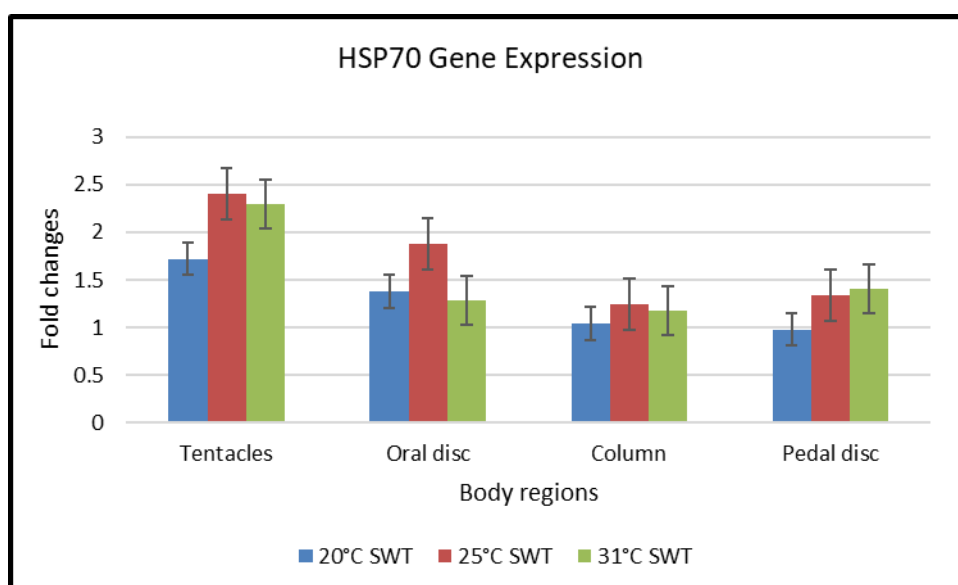


Figure 4.26: Variation of HSP70 gene expression in *U. claudenstina* body region (tentacle crown, oral disc, column and pedal disc) with increasing temperature

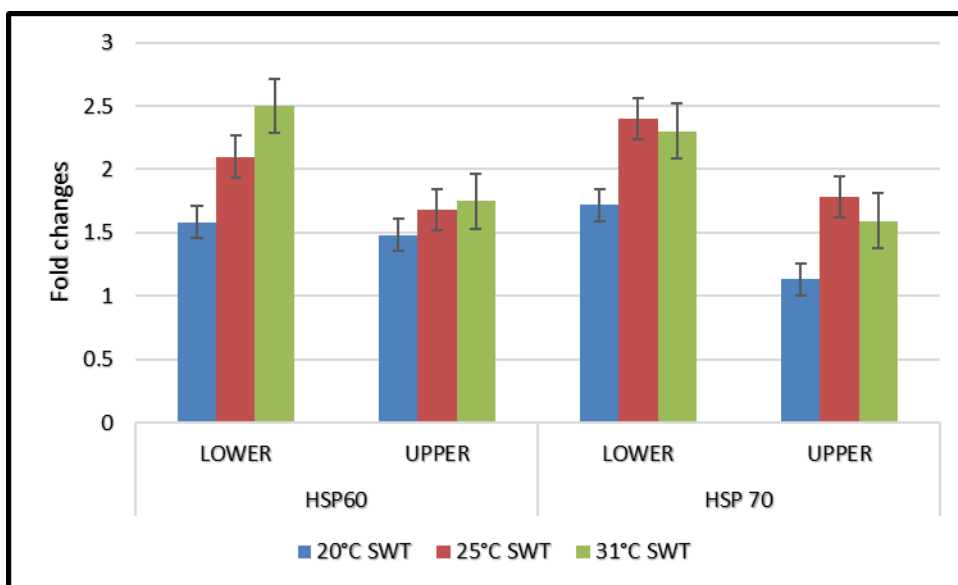


Figure 4.27: Variation of HSP60 and HSP70 gene expression in *U. claudeni* Tentacles with increasing temperature collected from upper and lower part of the tide pool.

Total 3 individuals were collected for each temperature. The benthic populations sampled in this study were collected from 3 and 5 m depth and experienced lower temperatures, despite being at lower latitudes.

We have examined the up-regulation of the HSP60 and HSP70 genes to assess the temperature tolerances. Results showed signal of expression of HSP60 and HSP70 gene in the different tissue of sea anemone (Tentacle, Oral disc, Column and Pedal disc). Differential expression of HSP60 and HSP70 was found in sea anemones according to seasonal changes in subtidal SWT (20°–31°C) along the Vadodra jhala site of Saurashtra coast. A significant level of HSP60 was recorded in specimens collected when the subtidal SWT reached 31°C, compared with the levels of expression when SWT ranged between 20°C and 25°C (Fig. 4.25-4.26). HSP70 shows higher expression with increase in temperature but HSP70 was less express than the HSP60 when temperature reaches to 31 °C. The Gene expression for HSP60 and HSP70 found higher in

the tentacle than the other tissue of sea anemone (Fig. 4.27). Nevertheless, significant differences were not found.

The heat shock response acts to protect organisms from possible damage to the cell protein resulting from a wide range of stressors including extreme temperature, osmotic stress, UV radiation and heavy metals. The response involves the rapid synthesis of a set of HSPs which allow the cell to withstand natural conditions. Therefore, in species that occupy stressful environments, the ability to produce this response is expected to play a significant role. HSP60 is a member of the chaperone family, known to bind target proteins under normal conditions to facilitate folding and assembly. The development of damaged proteins increases under adverse temperatures and induces a subsequent induction of HSPs, such as chaperones. The results of this study indicate that elevation of HSP60 levels is a phenotypic (acclimatization) adaptation of sea anemones to stressful SWT.

This study is well supported by findings related to the heat shock response of sea anemones during broad seasonal changes in sub-tidal SWT (Choreshe et al., 2001). It seems likely that 31°C is a stressful temperature for the sea anemones, necessitating a high expression of HSP60 for long periods (days to several weeks) for them to survive. The lower constitutive levels of HSP60 that occur at 20°–25°C may indicate that this HSP60 is less expressive when the sea anemone is under normal environmental conditions of the organism.

This work offers the use of HSP60 as a potential bio monitor, in addition to other HSPs such as HSP70 (Koziol *et al.*, 1996 and Pyza *et al.*, 1997), which will allow assessment of the impact of environmental changes on marine organisms. HSP70 proteins are necessary for protein folding, multimeric dissociation and association, translocation of proteins across membranes, and regulation of the heat shock response (Mayer and Bukau, 2005). The expression during thermal stress in of the HSP70 gene family was analysed in different tissue of sea anemone. The expression of HSP70 did not show a significant increase in tissues other than tentacles. The expression of the HSP70 in the tissues sampled tended to have a less gene expression compared

to the HSP60. It may be due to the other extreme abiotic factors prevalent in the natural habitat, (e.g., UV light, dissolved O₂) synergistically affected the expression of HSP60 and HSP70 in *Urticina claudeni*. Hence the observed expression of both genes in sea anemone in the field is the outcome of a combination of stressors, of which increased SWT is a major component.

These observations stress the importance of measuring both the HSPs response and the phenotypic parameters in order to fully understand the role of HSPs in organism responses to temperature stress and the cellular changes that occur from the initial stress response. In addition, we also emphasize the need for further in-depth studies that analyse sets of cellular biomarkers coupled with vision-based indices, such as degree of pigmentation, in order to monitor the sub-lethal cellular responses and the health.