INT. J. BIOL. BIOTECH., 14 (4): 561-573, 2017.

ULVA INTESTINALIS L.: MORPHOLOGY AND TEMPORAL VARIATION IN BIOMASS, THALLUS LENGTH AND REPRODUCTIVE PATTERN FROM KARACHI, PAKISTAN

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ABSTRACT

The morphology, thallus length, biomass and reproductive pattern of green seaweed *Enteromorpha intestinalis* (L.) Nees (now it is recognized as *Ulva intestinalis* L.) was studied from clean (Station1) and polluted (Station 2) sites in the Sandspit backwaters, Karachi. It was collected for seven months at St.1 and all the year around at St. 2 during the study period. The thalli at St.1 were light green, mature having minute holdfast whereas, at St.2 thalli were dark green, young , profusely branched, and studded with numerous hair-like branches, without holdfast found floating on water surface. The cells in young thalli were rectangular to polygonal in shape compactly packed in longitudinal rows with no sign of vegetative division and sporulation. In mature thalli the cells were polygonal showing vegetative division and sporulation. The chloroplast was parietal, cup-shaped with 1-3 pyrenoids at St.1 and 3-4 pyrenoids at St. 2. Variability in biomass, length, and cell diameters between populations of two stations was observed. At St. 1 sporulation was observed in most of the thalli and at St. 2 reproduction was by means of vegetative fragmentation throughout the year. At St. 1 blooms occurred during period of high sporulation whereas at St. 2 such blooms were common during peak of vegetative propagation.

Key words: Sandspit, young thalli, sporulation, blooms, *Ulva intestinalis.*

INTRODUCTION

The species of monostromatic green seaweeds *Ulva* belongs to family Ulvaceae. The species of *Ulva* have been reported as *Enteromorpha* initially, have short-lived filamentous algae with high nutritious value (Aguilera-Morales *et al*, 2005). They are preferred as food by many herbivorous animals such as fish, snails and wading birds (Lin *et al*., 2008). Several members of genus *Ulva* are also found fouling on ships (Boney, 1965), commonly on oil tankers (Evans and Christie, 1967) and are rapidly transported from temperate to tropical environment. Due to their tolerance to extreme environmental conditions, the successful settlement/establishment of zoospores has been observed (Biebl, 1962; Crisp, 1965). The life cycle of *U. intestinalis* showed haploid and diploid phases, which are morphologically similar. The proliferation of *U. intestinalis* in many parts of the thallus were found and it is the common feature of this species. The present study was carried out to study in detail the reproductive pattern of *U. intestinalis* from unpolluted and polluted sites and also to find out temporal variation in biomass and thallus length at two stations.

MATERIALS AND METHODS

Study area

Samples were collected from the backwater area of Sandspit (24° 85' N, 66°90' E) about 18 km Southwest of Karachi from two different stations (St.1 and St.2). Sandspit backwaters are comprised of shallow tidal lagoons, intertidal mud flats and mangrove swamps covering an area of about 400 hectares. The freshwater from Lyari River enters the backwaters from the eastern side and seawater from the south and from Karachi Harbor (Fig. 1).

The St.1 is located in the middle of mangrove forest near Wetland Centre having greater influx of seawater and considered as clean site. The mangrove *Avicennia marina* dominates the vegetation and forms nursery grounds for many species of marine fauna and flora. The St. 2 is located near Kakapir a small coastal village situated at the Western end of Sandspit backwaters. In the Kakapir area the polluted water through Lyari River rich in domestic sewage and industrial effluents pile up in narrow creeks because of poor flushing. Despite this a number of species of fauna and flora are of common occurrence (Fig. 1).

Sampling

The samples of *U. intestinalis* were collected from Station 1 during May 2011 to September 2011 and in January 2012 to February 2012 period. Whereas, at Station 2 they appeared during March 2011 to February 2012. For the estimation of biomass three quadrates (25× 25 cm) at an interval of 3m, horizontal to the shore were placed and the targeted species were scooped from substrate and collected in polythene bags. They were immediately brought to the laboratory where they were washed thoroughly squeezed with hand and placed on the blotting paper to remove excessive water. Wet weight of *U. Intestinalis* was recorded nearest to 0.01gm on electronic balance. The single thalli (N = 100) were spread on blotting paper and their lengths were measured nearest to 0.1 cm by using measuring scale. The lengths were recorded from the hold fast to the tip of frond in case of young thalli and from the base to the tip of the frond of mature thalli. The species was identified as described by Saifullah and Nizamuddin (1977) as *Enteromorpha intestinalis* which is presently known to be *Ulva intestinalis* L.

Macroscopic examination of thalli was carried out to study morphology, texture, color and, presence of branches in the basal region. A small portions of thalli were microscopically examined in surface view and thin transverse sections of *U. intestinalis* thalli were cut with a sharp razor blade, and temporary mounted on a drop of 100% glycerine on a glass slide to examine, cell morphology, sizes and arrangement; appearance of chloroplast and number of pyrenoids per cell. The cell sizes were recorded from the widest part of the cell.

Physical parameters

The physical factors, water and air temperature, salinity, pH and oxygen, were monthly recorded from both the sampling sites. Water and air temperature, and salinity were monitored *in situ*. The water temperature was measured by mercury thermometer, salinity by hand held refractometer (Atago, S/Mill-E) and pH by pH meter (Jenway Model 350 pH meter).Water samples for the estimation of oxygen after fixation by adding KI and $KMnO_4$ in stoppered glass bottles were brought to the laboratory and oxygen concentrations were recorded by Winkler's method described in Strickland and Parsons (1972).

RESULTS

Physical parameters

At St. 1 the average water temperature was 26 ± 5.0 °C and St.2 it was slightly higher (20 ± 2.0 °C). Salinity (36 ± 1.48‰), pH (8 ± 0.5) and dissolve oxygen (3.± 0.3 ml/L) concentrations were also somewhat higher at St. 2 as compared to St. 1 where these were 34 ± 2.0‰, 8.0 ± 0 and 3 ± 0.4 ml/L, respectively. The monthly variations in physical parameters of both the station are given Fig. 2.

Morphology

Young Thalli

Macroscopic observation

The young thalli of *U. intestinalis* of St. 1 and St. 2 were clearly distinguished by their colour and texture. At St.1 they were dark green in colour, firm, curved and thick in texture, attached to the substrate with several branches arising from the minute holdfast (Caespitose), few hair-like branches easily seen by naked eyes were present in the middle portion of the thalli. Though at St. 2 the thalli were dark green in colour but they had distinct black pigments and numerous hair-like branches (Fig. 3)

The young thalli at St. 1 were found throughout the sampling period except August. Their peaks were recorded in May 2011 (70%) and January 2012 (75%) at St. 1. Whereas at St. 2 they accounted 91.7 to 100% of total young thalli examined (Fig. 4). The average size of the young thalli was 29 ± 24 mm at St.1 and 65 ± 1 mm at St.2, respectively.

Microscopic observation

Microscopic observation of thalli revealed cell shape and arrangement. The surface view of the thalli showed mostly rectangular cells compactly packed forming distinct longitudinal rows, some cells were polygonal without any intercellular spaces. There was no sign of sporulation and vegetatively dividing cells. The cells contained cup-shaped parietal perforated chloroplast having 1-3 pyrenoids at St. 1 and 3-4 pyrenoids at St. 2 (Tables 1 & 2; Fig. 3).

Mature thalli

Macroscopic observation

On examination with naked eye the completely mature thalli both at St. 1 and St. 2 appeared to be yellow green in colour with delicate smooth surface (Fig. 3). At St. 1 they were attached to the substratum with a minute holdfast. Whereas, at St. 2 emerged as free floating, entangled masses, lacking holdfast on the surface of stagnant water. The mature thalli predominated throughout the year at St. 1 and their highest peak (100%) was recorded in August 2011. At St. 2 they constituted < 10% of thalli examined (Fig. 4). The average length of mature thalli was 77 ± 55.2 mm and 90 ± 3.0 mm, respectively at station St. 1 and St. 2.

Microscopic observation

At both the stations in surface view early maturing thalli though appeared to be dark green in colour but the cells of their middle portion contained spores. The cells of fully matured thalli were squarish, oblong or occasionally slightly polygonal, with rounded corners, loosely arranged radially, in a gelatinous matrix with distinct intercellular spaces. Most of the cells were filled with spores undergoing sexual reproduction. In mature thalli vegetatively dividing asymmetrical cells were also present. In sporulating cells the chloroplast was not clearly visible; however, in vegetatively dividing cells it was thread-like (Fig, 3).

Temporal variation in growth and reproductive pattern

Station 1.

Cross section of *U. intestinalis* showed monostromatic long thalli having single layer of girdle shape chloroplast with 1-3 pyrenoids. At the beginning of sampling in May to July period *U. intestinalis* thalli were dark green in colour, linear in shape and slightly curved at the middle. In most of the thalli in surface view the cells were quadrangular, polygonal to oval in shape while in early maturing thalli (30.0- 68.3%) the cells were quadrangular, compactly packed forming spores. In these months the mean cell size of thalli was 14 ± 20 to 27 ± 5.0µm found loosely embedded in gelatinous matrix, and hair-like branches observed all over the surface of thalli. In August the colour of some thalli changed to yellow green while mostly were dark green. They were fragile, slightly curved at the middle and hair-like branches covered the surface of the thalli. In August the mean cell size was 15 ± 3.3 µm. At the tip and in the middle portion of the thalli the cells were polygonal to oval and in the lower region of the thalli quadangular to polygonal in shape but not compactly arranged. Sporultion was visible in all cells except of the lower portion of the thalli that has no spores and many cells of thalli were translucent, after release of spores, remaining cells of thalli were asymmetrical undergoing vegetative cell division. In September the cell size increased to 23±3.3 µm, polygonal to oval in shape much loosely arranged in the entire thallus. In 90% of the thalli (mature thalli) the cells were translucent half-filled with spores (Fig. 3 & 5).

In the months of October to December 2011 the thalli of *U. intestinalis* were not found at St. 1. The young thalli of *U. intestinalis* started to appear in January 2012. They were dark green in colour and slightly firm, curved and minute hair-like branches were present in some thalli. In surface view cells of the upper region of the thalli were rectangular in shape, whereas, in the middle region they were rectangular and quadrangular showing asymmetrical division and were compactly packed in the lower region. The mean cell size was 13 ± 1.3 to 13 ± 2.3 µm, respectively, in January and February. In March also thalli were dark green, slightly firm and much curved but had few branches and mean cell size was 14 ± 20 µm. The mature thalli constituted 73.3%, and rests were young thalli. The cells appeared to be polygonal and quadrangular in shape with no sign of sporulation. In the upper region of thalli the cells were quadrangular in shape, loosely arranged in a gelatinous matrix, showing symmetrical cell division just below the tip of the thalli. While in the middle region of thalli the cells were polygonal, dividing asymmetrically and in lower region cells were rectangular and compactly arranged (Fig. 3 & 4). In T.S the cells appeared to be arranged in single row, were rectangular with rounded corners. Significant difference in cells diameter was recorded at St. 1 ($F = 75.5$, $P < 0.05$).

Station 2.

Majority of thalli of *U. intestinalis* at St. 2 were dark green in color and dark blackish pigments were visible in surface view. They were found throughout the year, mostly showed vegetative reproduction and only few thalli were observed producing spores. During March 2011 to February 2012 period only 3.3 to 8.3% thalli were yellow green in colour. In the study period the cells in dark green thalli were quadrangular in shape containing 2-4 daughter nuclei. Whereas, in yellow green thalli which were found in March, May and December the cells were polygonal in shape containing spores. The cell diameter was 14.2 to 17.1 µm in this period except for February 2012 when their size increased to 23.4 µm (Fig. 3-5). The difference in cell diameter in different months was weakly significant ($F = 10.02$, $P<0.05$.)

Fig. 1. Showing sampling site Sandspit Station 1 (St. 1) and Station 2 (St. 2).

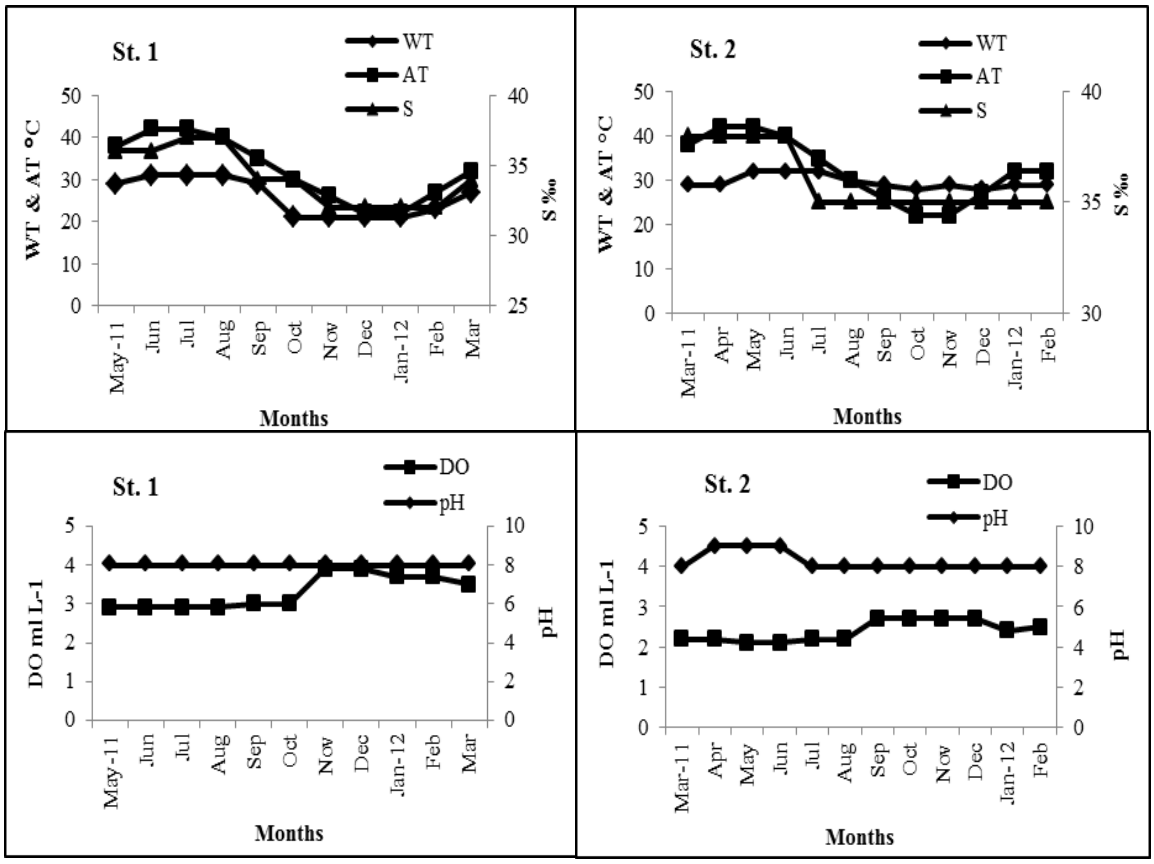

Fig. 2. Monthly variation in watertemperature (WT), air temperature (AT), salinity (S), dissolvedoxygen (DO) and pH at St.1 and St. 2.

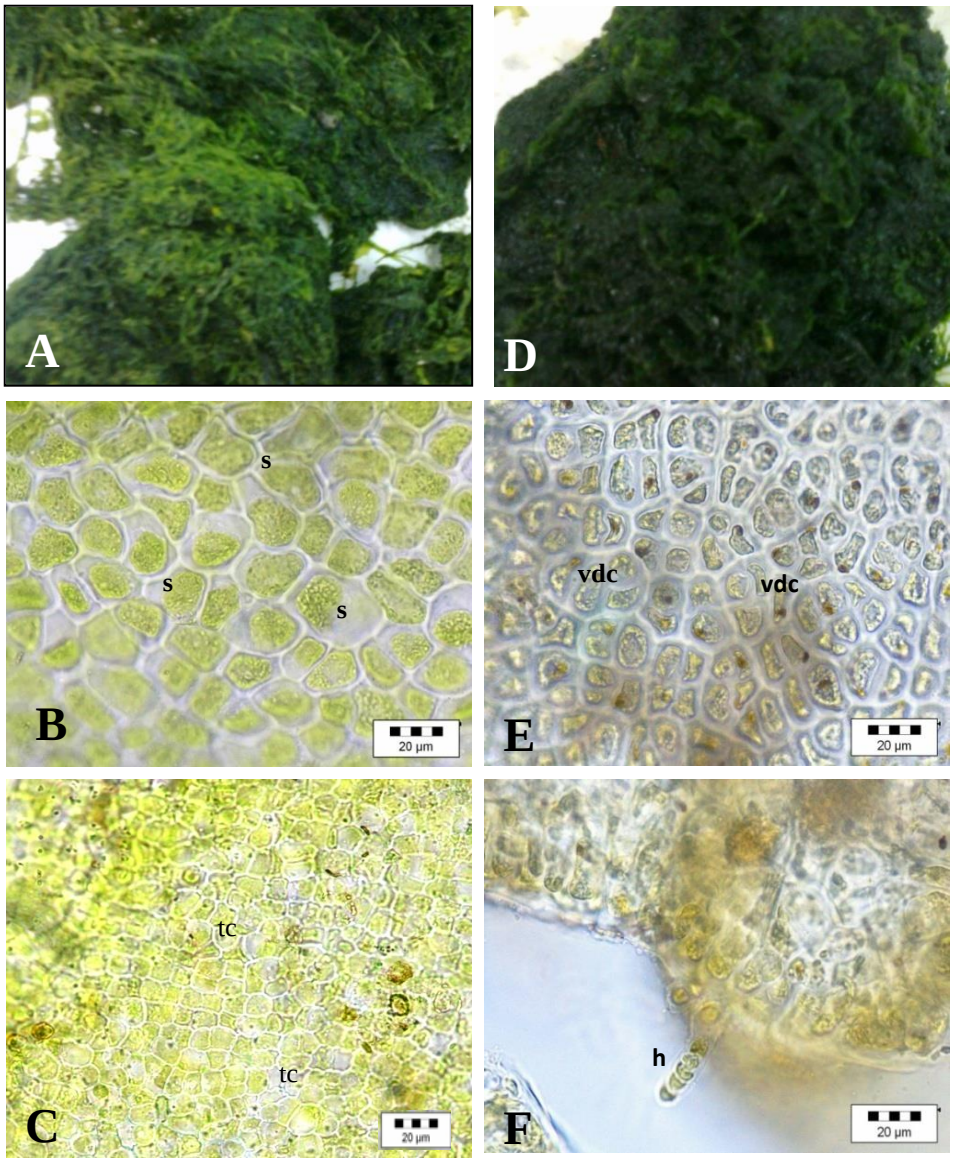

Fig. 3. *U. intestinalis:* St. 1. A- yellowish green thalli, B- Surface view of thalli showing sporulated cells, C- translucent cells after release of spores. St. 2. D- dark green thalli, E- vegetativly dividing cells, F- development of hair-like branches. Abbreviations: s = spores, tc = translucent cells, vdc = vegetative dividing cells, h = hair-like branch.

Temporal variation in biomass

Station 1.

The biomass of *U. intestinalis* in different months at this site was found to be significantly different (F=46.20, P< 0.05). At the beginning of sampling period in May 2011, the biomass of *U. intestinalis* was 208.33 ± 2.9 g^{-m2}, reached the, peak in June (210 ± 5.0 g^{-m2}) with profuse sporulation of fronds. From July onward biomass of *U. intestinalis* gradually decreased from 150 ± 20.0 g^{-m2} in August to 128±10.4 g^{-m2} in September after the release of spores. During October to December *U. intestinalis* completely disappeared at this site and again reappeared in January 2012, with a biomass of 170 ± 5 g^{-m2} which increased to 205 ± 5.0 g^{-m2} in February and more or less similar biomass persisted in March, and April 2012 (Fig. 6). The biomass in different months was found to be significantly different (F = 46.2, *P* <0.05).

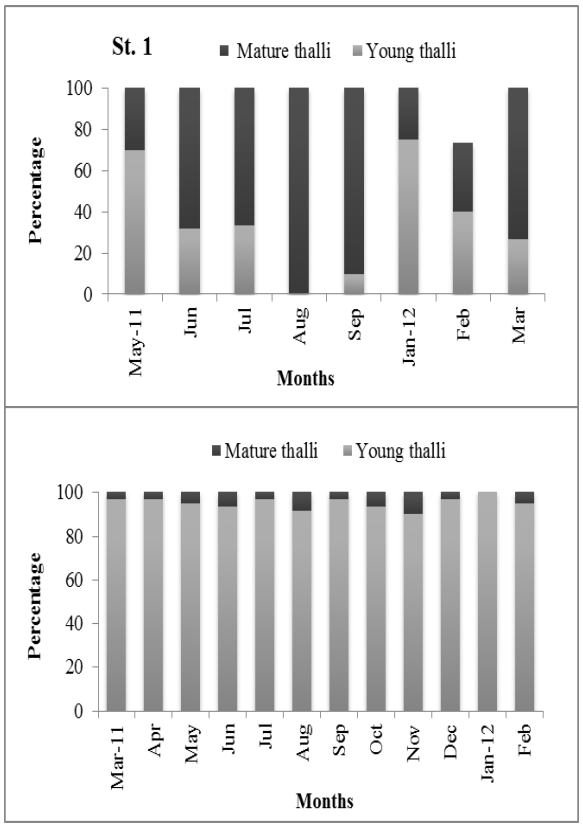

Fig. 4. *U. instestinalis:* monthly variation in occurrence of mature and young thalli at St. 1 and St. 2.

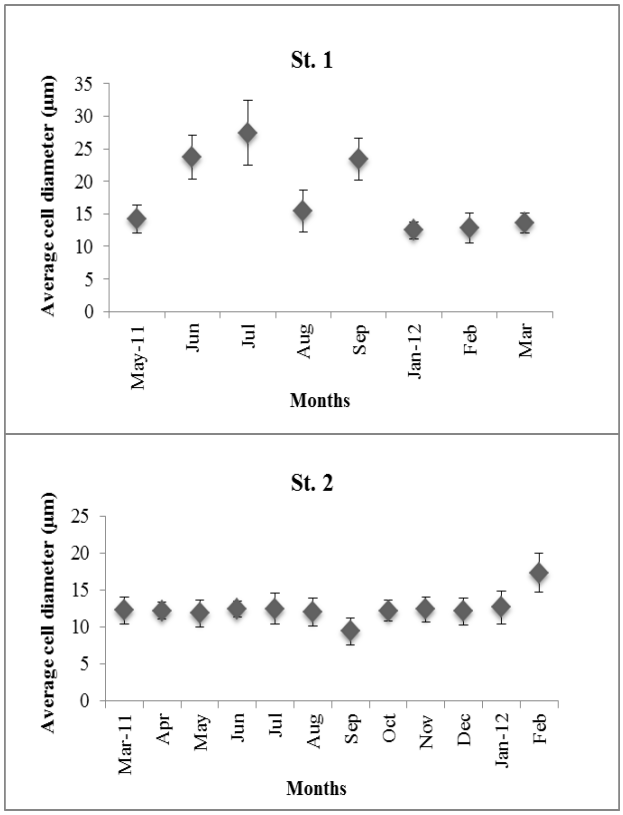

Fig. 5. Temporal variation in cell diameter of *U. intestinalis* at St. 1 and St. 2. Bars showing standard deviations.

Station 2.

In the beginning of sampling period in March 2011 at St. 2 the biomass was 200 ± 1.0. g^{-m2} and in May 2011minimum biomass (138. g^{-m2}) was recorded. However, in the period June 2011 to February 2012 it was in the range of 201 ± 4.0 to 240 ± 10.0 g^{-m2} (Figure 6). One way ANOVA showed significant monthly difference ($F=37.9$, $P<0.05$) in biomass of seaweed.

Temporal variation in length frequency

Station 1.

At Station1 during May to September 2011 the length frequency of *U. intestinalis* was. 54.0 ± 32.65 to113.15 ± 12.50 mm, respectively. However, in October to December 2011 period *U. intestinalis* disappeared from sampling site. In January 2012 they reappeared and their average length was 49.2 ± 13.7 mm and it markedly increased to 104.9 ± 51.1 mm March 2012 (Fig. 7).The monthly lengths of thalli were significantly different ($F=38.2$, $P < 0.05$).

Station 2.

At Station 2 which is the polluted site *U. intestinalis* grows throughout the year. In March 2011 average length was 63.2±18.2mm and it gradually increased to 75.5 ± 10.2 mm October to reach the maximum size in November. However, in December 2011 to February 2012 their average length was 64.5 ± 13.1to 65.6 ± 12.5 mm (Fig. 7). The lengths of thalli were significantly different in the study period ($F =4.4$, $P < 0.05$).

Correlation between mean length of thalli and biomass

At Station 1 the relationship between mean length of thalli and biomass showed moderate correlation ($r^2 = 0.37$; $p<0.05$) whereas, at St. 2 the correlation was non significant ($r^2= 0.01$; $p>0.05$).

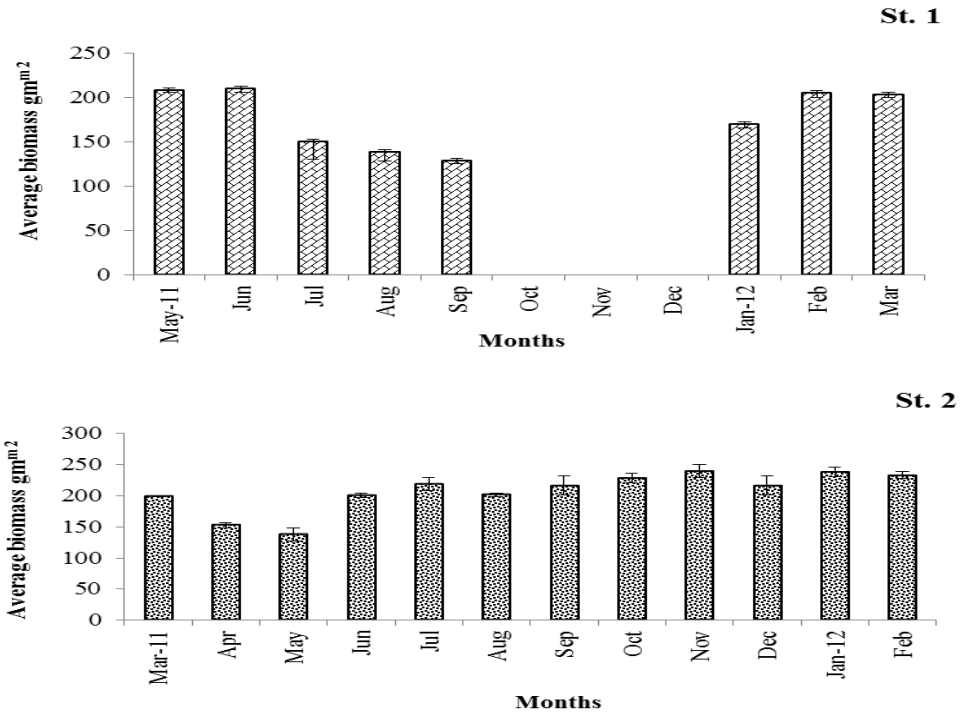

Fig. 6. Temporal variation in biomass of *U. instestinalis* at St. 1 and St. 2. Bars showing standard deviations.

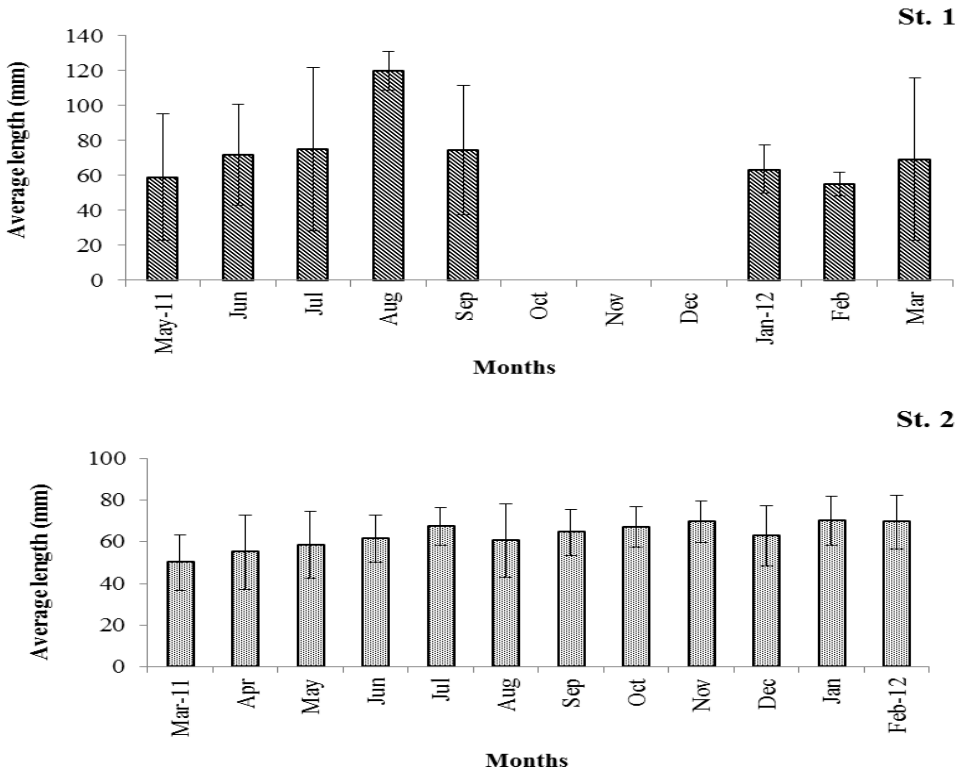

Fig. 7. Temporal variation in length frequency of *U. instestinalis* at St. 1 and St. 2.Bars showing standard deviations.

Table 1. Morphometric characteristics of *Ulva intestinalis* from St.1 (Unpolluted site).

Month	**hape**	**Thallus colour**	**Cell shape**	**Cell diameter (µm)**
May-11	Slightly curved at the middle, fragile, hair like branches present	Dark green	Cells of middle regions were quadrangular, polygonal and loosely embedded in the gelatinous matrix. Cells of the middle portion of some thalli containing spores, rest of the cells were undergoing symmetrical and asymmetrical vegetative division.	14.18±2.14
Jun	-do-	-do-	Cells of middle region were loosely embedded in gelatinous matrix, quadrangular, polygonal to oval in shape. In the middle portions of thalli vegetatively dividing asymmetrical daughter cells present, in few thalli cells contained spores. The lower portion of thalli containing compactly packed cells with no sign of cell division. In upper region cells were quadrangular.	23.72±3.41
Jul	-do-	-do-	Cells loosely embedded in gelatinous matrix, quadrangular to oval in shape, middle portion of thalli showing few vegetatively dividing asymmetrical daughter cells. The cells in middle mostly containing spores. Cells in the upper and lower portion were quadrangular and rectangular, some what loosely arranged showing asymmetrical and symmetrical cell division.	27.43±4.98
Aug	-do-	Dark green to yellow green	Cells at the tip and middle portion of thalli were polygonal to oval in shape, whereas, in the lower region cells were quadrangular and rectangular but not much compactly arranged, Sporulation visible in all cells, except in the lower portion of thalli that has no spores but vegetatively dividing cells were present.	15.43±3.26
Sep	-do-	-do-	Cells of thalli oval to polygonal in shape, loosely arranged, mostly translucent after release of spores. However, few cells partially filled with spores.	23.41±3.25
Jan-12	Entire thallus curved, firm, few minute branches present in some thalli.	Dark green.	Cells rectangular in shape, compactly arranged. No vegetatively dividing cells and sporulation visible. Cells in the lower region of thalli were more compactly packed than the middle region.	12.49±1.29
Feb	-do-	-do-	In upper region of thalli cells rectangular to oval in shape. While in the middle region rectangular and quadrangular, showing slightly loose arrangement undergoing asymmetrical cell division. In the lower region cells rectangular and compactly packed.	12.79±2.32
Mar	Entire thallus curved, firm, few hair-like branches with greater number of marginal teeth in some thalli.	-do-	Cells in the upper region of thalli were quadrangular loosely arranged, undergoing symmetrical cell division just below the tip of thalli. In the middle region cells polygonal, dividing asymmetrically. In the lower portion cells rectangular and compactly packed.	13.66±1.53
Apr	-do-	-do-	In upper and mid region quadrangular, polygonal loosely arranged cell present showing asymmetrical and symmetrical cell division. In lower region cells rectangular in shape	23.46±4.07
May	-do-	Dark and light green	In upper and mid region cells quadrangular and polygonal, loosely arranged with few spores in middle portion cells. In lower region cells showing asymmetrical and symmetrical cell division, quadrangular in shape.	24.44±3.19
Jun	-do-	Light green	In upper and mid region quadrangular, polygonal loosely arranged cell, spores found in middle portion. In lower region cells undergoing asymmetrical and symmetrical cell division quadrangular in shape.	15.83±2.68
Jul	-do-	-do-	In upper and mid region cell quadrangular, polygonal loosely arranged, spores found in upper and middle portion of thalli. In lower region asymmetrical and symmetrical quadrangular cells were found.	10.49±2.20

Table 2. Morphometric characteristics of *Ulva intestinalis* from St.2 (Polluted site).

Month	**Thalli Structure**	**Thallus color**	**Cell Shape**	**Diameter µm**
Mar-11	Fragile, slightly curved in the middle, upper and lower parts more curved. Hair-like branches present which are of large size in the middle of the thallus and in the lower region small marginal teeth present.	Dark green	Cells rectangular, in the lower region middle cells dividing vegetatively but in few thalli cells of middle portion polygonal in shape, partially filled with spores.	12.26±1.83
Apr	-do-	Dark green	-do-	12.20±1.15
May	-do-	Dark green	Cells polygonal in shape in the upper region.Cells of the middle and lower region quadrangular. In the middle region cells asymmetrical vegetatively dividing. But in few thalli which appeared to be damage the cells of entire thallus polygonal in shape and partially filled with spores.	11.86±1.82
Jun	-do-	Dark green	-do-	12.40±1.07
Jul	-do-	Dark green	Cells of entire thallus rectangular, vegetatively dividing, but in few thalli cells were polygonal and translucent.	12.47±2.07
Aug	Slightly curved at the middle, fragile, few hair-like branches in the middle region.	Dark green	-do-	12.07±1.86
Sep	-do-	Dark green	-do-	9.46±1.82
Oct	Slightly curved in the middle, fragile and smooth. Long and small hair-like branches observed at the margins of the entire thallus with middle region predominated by long teeth.	Dark green	-do-	12.24±1.41
Nov	-do-	Dark green	-do-	12.38±1.74
Dec	-do-	Dark green	Cells rectangular in the lower region and middle region cells undergoing vegetative division in most of the thalli. In few thalli cells polygonal in shape partially filled with spores.	12.12±1.82
Jan-12	-do-	Dark green	Cells of entire thallus rectangular to quadrangular, undergoing vegetative symmetrical and asymmetrical cell division No sporolated or translucent cells present.	12.68±2.25
Feb	-do-	Dark green	-do-	17.34±2.60

Blooms

At Station 1 which is a clean site, sporulated thalli of *U. intestinalis* were found attached to the rocks. Blooms occurred in August to September period when sexual reproduction was intense. In these months thalli dispersed their spores that spread over an area of approximately 1 km long strech. Such blooms were also noted in May and June when maximum biomass was noted with very little sporulation but pronounced fragmentation in thalli, resulted in the formation 1-2 m long dense patches spread over an area of approximately 2 km long and 25 m wide.

At St.2 the 2-3 meter long dense patches of monostromatic thalli of *U. intestinalis* were found spread over an area of 2-3 km long and 1 km wide unattached to the soil in July, October, November 2011 and January 2012. They were found to reproduce asexually as a result of fragmentation and regeneration throughout the year causing blooms.

DISCUSSION

This species was earlier identified as *Enteromorpha intestinali*s. The molecular analysis of this species was carried out and it was revealed that *E. intestinalis* and *Ulva intestinalis* are same species (Tan *et al*., 1999; Hayden *et al*., 2003) therefore in the present study it is designated as *U. intestinalis*.

In the present work revealed the reproduction, morphology and biomass of *U. intestinalis.* It has been studied from two stations St. 1(unpolluted) and St. 2 (polluted). Morphological observation showed variability in coloration and branching of thalli at two stations. At St. 1 the coloration of thalli was yellow to dark green with few branches present. The specimens procured from St. 2 were dark green in colour, numerous branches and black spots were present on the surface of thalli. St. 2 is located at a polluted site receiving domestic, industrial sewage and effluents from port and harbor (Manzoor *et al*., 2011; Nergis *et al*., 2012). It is a well-known fact that morphological changes in algae are usually associated with altered nutrient supply in eutrophic condition, changes in salinity and increased photon irradiance (Valiela *et al.,* 1997; Reed and Russel, 1978). Therefore, profuse branching in *U. intestinalis* at St. 2 supports the view that abnormal growth forms which are one cell thick facilitate nutrient uptake and provide high surface-to-volume ratio (Valiela *et al*., 1997), because of this species of *Ulva* are the main component of green tides in various parts of the world (Poole and Raven, 1997; Blomster *et al*., 2002). On the coast of Finland they become free-floating in the water column forming extensive blooms (Back *et al*., 2000). The variability in color of thalli at two sites is due to variability in nutrient concentration and mode of reproduction (Neori 1996, Neori *et al*., 1996, Neori *et al*., 2004). This is also supported by the fact that *U. intestinalis* at St.1 were observed as attached form with minute hold fast, reproducing mostly by sporulation giving rise to attached form which later on reproduce vegetatively in few months whereas, at St. 2 they were found free floating on the surface of water without any hold fast, otherwise similar to attached form, mostly reproducing asexually and very few sporulating thalli were observed. Previously it has been reported that floated forms arise as a result of vegetative reproduction or regenerative growth (Hiraoka, 2003; Zhang *et al*., 2009). Presence of branches and scarce sporulation in this species indicate that at St.2 this species prefer vegetative or asexual mode of reproduction as reported by Lin *et al*. (2008) from Yancheng City China.

As *U. intestinalis* at St. 1 and St. 2 exhibit different mode of reproduction therefore, the variability in length of thalli was observed. However, at both stations the correlation between length of thalli and the biomass was found to be statistically insignificant ($p>0.05$). At St. 1 when sporulation was on peak in August the greater length of thalli was noted and the biomass was low. During October to December period *U. intestinalis* completely disappeared after release of spores, representing dependence of biomass on sporulation. Similarly, release of spores and gametes caused the loss of vegetation, reduction in length, biomass and disappearance of vegetation in *Ulva fasciata* for certain months in India and Mexico (Subbaramaiah, 1970; Grijalva-Chon *et al*., 1985). *U. intestinalis* at St. 2 was found throughout the year as vegetative free floating thalli due to nutrient enrichment as a result of pollution from sewage and poor flushing because of low wave action. Previous reports indicate that the growth and development of opportunistic green seaweeds increases in eutrophic conditions (Kraufvelin *et al*., 2009; Fatemi *et al*., 2012).

Two genera of Ulvophyceae, *Enteromorpha* and *Ulva* particularly are known to cause majority of blooms and are most common fouling algae (Fletcher, 1996; Stanley *et al*., 1999; Callow *et al*., 2000). In the present study the blooms of *U. intestinalis* were of greater occurrence at St. 2 which is a polluted site. Similarly, in areas polluted with domestic sewage, fertilizers and nitrogen rich water green tides have been observed recurrently (Fletcher, 1996; Hermandez *et al*., 1997; Wang *et al*., 2008). At St. 2 the length of thalli and biomass of *U. intestinalis* was more, or less constant throughout the year. Most of the thalli were reproducing vegetatively and very few were seen sporulating. Frequent blooms of this species were recorded at this site during the study period. Zhang *et al*. (2009) have reported that vegetative growth of *U. prolifera* causes greater blooms as compared to sporulation (Dayton 1985; Reed *et al*., 1988; Hoffman and Santelices, 1991). Sheet-forming of *U. intestinalis* both in field and laboratory

culture reproduce by spores and blooms in summer by fragmentation (Poole and Raven, 1997; Valiela *et al*., 1997; Reed and Russell, 1978; Blomster *et al*., 2002). At Veerse Meer, Netherland free-floating green tide *U. scandinavica* very seldom reproduce by spores instead they over winter buried in sediments (Kamermans *et al*., 1998; Malta *et al*.,1999). Thalli of *U. prolifera* that are buried in sediment can cause bloom because somatic cells have the tendency to regenerate faster under stress conditions and produce blooms which can last for two months and with stand changes in temperature (Zhang *et al*., 2009). Furthermore, spores can travel only few meters, but prolific vegetative cells can cover greater distances because they have tendency to tolerate stress (Sousa, 1984; Hoffman, 1987; Santelices, 1990). On the contrary green tides and attached populations of *U. armonican* in Brittany, Francere produce by spores (Coat *et al*., 1998). Somatic cells have greater tendency to resist predations as compared to spores which are susceptible to predation (Dayton, 1985; Reed *et al*., 1988; Hoffman and Santelices, 1991).

The specimens of St.1 and St.2 showed no variability in cell shape. The young thalli of both sites had rectangular cells arranged in longitudinal rows while the mature thalli showed polygonal to oval cells with radial arrangement. In recent study on young and mature thalli of *E. flexuosa* the shape of cells and their arrangement was found to be different. In young thalli they were rectangular in shape and their rows were distinct whereas, in mature thalli the cells were less regular and the rows were not distinct (Messayasz *et al*., 2013).

The mean cell sizes in the present study at St. 1 were in the range of 12.49 ± 1.29 to 27.43 ± 4.98 µm and St. 29.46 ± 1.82 to 17.34 ± 2.60 µm quadangular to polygonal in shape. Saifullah and Nizamuddin (1992) reported10-20 µm high and 15 µm broad single layer of rectangular cells, parietal chloroplast with 1-2 pyrenoids in *E. intestinalis* from Jeddah, Saudi Arabia. Blomster (2002) reported from Finland in bloom forming fragile sheets of *E. intestinalis* the cells were 5-4 µm diameter, irregularly rounded to elongate having hood-shaped chloroplast and 1-2 pyrenoids. When callus like tissue from these sheets were grown in laboratory condition it resembled *U. intestinalis* tubular thalli having rectangular to rounded-polygonal cells arranged in short rows with 1-3 pyrenoids but they never branched and developed into monostromatic sheets.

According to recent research this species of enteromorpha belong to genus Ulva on the bases of genetical observation made by (Tan *et al*., 1999). It is first time reported from the coast of Karachi, Pakistan.

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(*Accepted for publication September 2017*)