

# Macro and micro nutrients of seagrass species from Gulf of Mannar, India

## Abstract

The present study determined the macro (Phosphorus-P, Potassium-K, Calcium-Ca, Magnesium-Mg, Sodium-Na) and micro (Zinc-Zn, Iron-Fe, Nickel-Ni, Lead-Pb, Chromium-Cr, Cadmium- Cd and Copper-Cu) nutrients and proximate composition of protein, lipid, carbohydrate, chlorophyll, ash and fiber in three seagrass species namely, *Cymodocea serrulata*, *Halodule uninervis* and *Syringodium isoetifolium* from Tuticorin group of Vann island from Gulf of Mannar. The results show that the macro and micro nutrients and proximate composition vary with the species. Protein, carbohydrate, chlorophyll and fiber were high in *Cymodocea serrulata* with a composition of 19.1, 19.8, 2.12 and 37% respectively. Lipid and ash contents were high in *Halodule uninervis* with a composition of 5.3 and 28.7% respectively. All the proximate compositions were low in *Syringodium isoetifolium* compared with the other two seagrass species. Evaluation of mineral content demonstrates that the concentration of phosphorus, potassium and magnesium were high in *Cymodocea serrulata* (126, 615 and 1015mg/100g respectively), sodium was comparatively high in *Halodule uninervis* (1019mg/100g) and calcium was high in *Syringodium isoetifolium* (690mg/100g). Micronutrients such as zinc ranged from 22.21 to 38.36µg/100g, iron from 540 to 1074µg/100g, copper from 0.75 to 7.11µg/100g, nickel from 6.35 to 12.3µg/100g and lead from 1.22 to 7.5µg/100g. Chromium and cadmium were below the detectable level in *Halodule uninervis*. The results suggest that all the seagrasses have a great nutritional value and could be used as excellent nutritional supplements for the aquatic animals. The findings on nutrients composition of the seagrass in this study can be further used as a basis to study the assimilation in the aquatic animals, to prepare the nutritional information guideline and for other advanced research on seagrass.

**Keywords:** nutritional composition, seagrass, micronutrients, macronutrients

Volume 6 Issue 4 - 2018

Immaculate Jeyasanta K, Lilly TT, Jamila Patterson

Manonmaniam Sundaranar University, India

**Correspondence:** Immaculate Jeyasanta K, Suganthi Devadason Marine Research Institute, Manonmaniam Sundaranar University, Tirunelveli, Tamil Nadu, India, Email [immasdmri@gmail.com](mailto:immasdmri@gmail.com)

**Received:** July 27, 2018 | **Published:** August 07, 2018

## Introduction

Seagrasses are submerged marine angiosperms. They produce flowers, fruits and seeds and have roots, leaves and underground stems (rhizomes), which enable them to form an extensive network below the surface of the water.<sup>1</sup> They are primary producers and play a key role in the marine ecosystems.<sup>2</sup> Their beds purify water, cycle nutrient, stabilize sediment and dampen waves and underwater currents.<sup>3,4</sup> They also act as a “carbon sink” by absorbing carbon dioxide from the atmosphere. This, in turn, helps to slow down the effects of global warming. On the other hand, seagrasses provide food for a wide array of species, including, manatees, sea turtles,<sup>5</sup> sea urchins, waterfowl, gar and pinfish. They are also considered as a source of food production for man as they serve as a nursery ground to juvenile stages of economically important species of finfish, oysters, clams and shellfish.<sup>4</sup> Seagrasses have a positive influence on the health of marine animals that feed them. Turtles consuming seagrasses grow faster, attain sexual maturity earlier and the females produce more eggs<sup>6</sup> compared to that of other turtles present in the wild. This is attributed to the high protein content and easily digestible nature of seagrasses. Thus, seagrass would play a major role in the long-term viability of the species.<sup>7,8</sup>

The global diversity of seagrass species is low. There are only 12 genera with 58 species of known seagrasses.<sup>9</sup> Seagrasses can be found

distributed from the mid intertidal areas to depths greater than 50m. In India, 14 species of seagrasses have been recorded along the east and west coasts.<sup>10</sup> Gulf of Mannar, Palk Bay, Andaman Nicobar Islands and Lakshadweep Islands are known for their seagrass resources. Seagrass species *Cymodocea serrulata* and *Syringodium isoetifolium* contribute more biomass in these regions and their photosynthetic productivity is also higher compared to the other ecosystems of the world.<sup>11</sup> They are used as “classical” bio-indicator species providing many valuable ecological and economical services within coastal and estuarine areas.<sup>12</sup> Global distribution of seagrass genera is remarkably consistent in the northern and southern hemispheres, sharing ten seagrass genera and each hemisphere has only one unique genus. Some genera are much more spacious than others.<sup>13</sup> It is well accepted that seagrasses affects the chemical, physical and biological environment significantly, and is therefore considered as an “ecological engineer”. Seagrasses not only inhabited deposited substrates but also play an important role in trapping sediments and stabilize them with vast root mat and at the same time absorb nutrients from the sediments to flourish both seagrass beds and their interrelated environment.<sup>14</sup> According to Malea,<sup>15</sup> seagrasses uptake high concentrations of potassium, magnesium, calcium, iron and sodium, which take part in the physiological processes of aquatic plants. The uptake of these nutrients is influenced by the nutrient concentrations in the sediments and dissolved nutrients in the seawater. Although seagrasses are widespread both in temperate and tropical areas, they do not adapt

to the change in chemical, physical and biological environment. They may have different optimal requirements based on the climate regimes.

Studies on the variation of multi-nutrients, especially trace elements, in the tropical seagrasses have been carried out by, Malea et al.,<sup>15,16</sup> Kannan et al.,<sup>17</sup> Kilminster<sup>18</sup> and Thangaradjou et al.<sup>19</sup> However, most of these studies are in Europe and the study on the nutritional value of seagrass in this study area is scarce in literature. The rapidly expanding scientific knowledge on seagrasses has led to a growing awareness that seagrasses are a valuable coastal resource. A healthy condition of seagrass meadows is detected through the nutritional value. Nutritional value of sea grasses are poorly studied compared to coral reef and mangrove forest. Through there is much focus on research on the seagrasses of the world over the last two or three decades, studies on their biochemical composition and their possible utilization as the substitutes for the existing food sources are still scanty. Present study was conducted to find out the proximate composition (macro nutrients) and the micronutrients content of the dominant seagrass species of the Gulf of Mannar, so as to evaluate them for use as food for human consumption.

## Materials and methods

Three species of fresh seagrasses were collected by hand picking method. The samples were washed thoroughly by using seawater to remove epiphytes, sand and debris. They were placed in food-grade plastic bags and transported to the laboratory in insulated containers. The samples were identified to species level based on the examination of morphological and anatomical characteristics and using taxonomic.<sup>1</sup>

In the laboratory, the fresh seagrasses were thoroughly rinsed with filtered seawater for three times. The remaining epiphytic algae, invertebrates, sand and debris were removed by hand. Leaves and rhizomes of individual species were separated. The fresh leaves were taken for chlorophyll estimation. All the rest portions were placed on aluminum foil trays and dried at 60°C in a hot air oven for a week to attain a constant weight. The dried samples were then grounded into a fine powder (to pass through a 1mm sieve) using a grinder and stored in air-tight labeled glass jars and placed in a desiccator at room temperature. The desiccator maintains the relative humidity below 5.5%. All chemical analyses were conducted in triplicate on the dried ground material.

## Measurements of components

The compositions of macro nutritive components were measured using standard procedures listed in Table 1.

**Table 2** Macro elements content of seagrasses

Macro minerals (mg/100g)	<i>Cymodocea serrulata</i>	<i>Halodule uninervis</i>	<i>Syringodium isoetifolium</i>
Phosphorus	126±1.0 <sup>c</sup>	110±1.0 <sup>a</sup>	118±1.12 <sup>ab</sup>
Sodium	978±1.05 <sup>b</sup>	1019±2.65 <sup>c</sup>	272±1.0 <sup>a</sup>
Potassium	615±5.0 <sup>c</sup>	320±2.0 <sup>b</sup>	149±1.0 <sup>a</sup>
Calcium	600±1.0 <sup>a</sup>	600±10.0 <sup>a</sup>	690±2.65 <sup>b</sup>
Magnesium	1015±2.65 <sup>c</sup>	920±8.66 <sup>ab</sup>	918±2.65 <sup>a</sup>

Data are mean values±S.E in rows with different superscript alphabet are significantly different (p<0.05) DMRT.

**Table 1** Procedures used for analysing various biochemical

Sl. No.	Biochemical	Standard procedure
1	Protein	Protein by Folin Reaction using a salt-free bovine serum albumin as a standard <sup>20</sup>
2	Lipid	chloroform- methanol method <sup>21</sup>
3	Total Carbohydrate	Phenol sulphuric acid method <sup>22</sup>
4	Fiber	Sulfuric acid and sodium hydroxide solution with ignition <sup>23</sup>
5	Ash	By Muffle Furnace <sup>24</sup>
6	Total chlorophyll	Phytochemical methods of testing Using 80% acetone <sup>25</sup>

## Digestion of seagrass for micro nutrients analysis

The seagrass samples (0.2g) were digested in nitric and sulphuric acids in a ratio of 0.5:1w/w basis. The mixture was heated at 80°C for 4hours on a hot plate. The digest was then filtered through 0.45µm Millipore filter and diluted with 50ml deionised water and analyzed for macronutrients (N, P, K, Ca and Mg) and micronutrients (Fe, Zn, Mn and Cu) using an atomic absorption spectrophotometer equipped with a hollow cathode lamp for each element and an air-acetylene burner against mineral elements standards. Determination of phosphorus by the molybdenum blue method using spectrophotometer at 720nm.<sup>24,26</sup>

## Statistical analysis

Individual differences between species were determined by Duncan's multiple range tests using SPSS/PC- package. The analyzed data were expressed as mean±standard deviation (SD). Numbers within the same rows followed by a different superscript alphabet were significantly different (P=0.05, DMRT).

## Results

The biochemical constituents of the three seagrasses namely, *Cymodocea serrulata*, *Halodule uninervis* and *Syringodium isoetifolium*, were shown in Table 2. The result showed that protein content in *C.serrulata* (19.1%) was higher than that of *H.uninervis* (13.6 %) and *S.isoetifolium* (12.8%). The Chlorophyll content ranged from 1.36 to 2.12%, while the carbohydrate and lipid contents of *C.serrulata* (19.8, 4.8 %), *H.uninervis* (13.24, 5.3%) and *S. isoetifolium* (14.25, 5.02%). All the species contained <10% crude lipid. Ash and fiber contents were the most abundant component in the dried material and were 24.2 and 37% in *C. serrulata*, 28.7 and 34% in *H. uninervis* and 18.2 and 27% in *S. isoetifolium*. Data analyzed by DMRT test showed numbers within the same column followed by a different superscript letter were significantly different (P=0.05).

Table 3 represents the concentrations of major elements of the three seagrass species. The results showed great variations of these elements. The maximum value of magnesium was recorded in *C. serrulata* (1015mg/100g) followed by *H.uninervis* (920mg/100g) and *S. isoetifolium* (918mg/100g). On the other hand, a same value of calcium was observed in *C. serrulata* and *H. uninervis* (600mg/100g), while the calcium composition in *S. isoetifolium* was 690 mg/100g. Sodium and potassium elements in *C. serrulata* were 978 and 615mg/100g and in *H. uninervis* they were 1019 and 320mg/100g, whereas *S. isoetifolium* had 272 and 149mg/100g respectively. The maximum value of 126mg/100g of phosphorus was recorded in *C. serrulata*, whereas 110 and 118mg/100g were observed in *H. uninervis* and *S. isoetifolium* respectively. Data analyzed by DMRT test showed numbers within the same column followed by a different superscript letter were significantly different (P=0.05).

The concentrations of microelements of all three seagrass species were shown in Table 3. Copper was below the detection limit in *C. serrulata* and *S.isoetifolium* species, whereas in *H. uninervis* 4.36µg/100g was recorded. In *S. isoetifolium*, zinc was higher (38.36µg/100g) than lead (7.5µg/100g) and nickel (9.18µg/100g). The highest iron content was found in *S. isoetifolium* (1074µg/100g) followed by *H. uninervis* (945µg/100g) and *C. serrulata* (540µg/100g). Chromium was below the detectable level in *H. uninervis* and *S. isoetifolium*, and cadmium was below the detection limit in *H.uninervis*. The lowest concentrations of chromium and cadmium (0.028 and 0.03µg/100g) were recorded in *C. serrulata* and only cadmium was found in very low concentration in *S. isoetifolium* (0.01µg/100g). Data analyzed by DMRT test showed numbers within the same rows followed by a different superscript letter were significantly different (P=0.05).

**Table 3** Micro elements content of seagrasses

Micro minerals (µg/100g)	<i>Cymodocea serrulata</i>	<i>Halodule uninervis</i>	<i>Syringodium isoetifolium</i>
Copper	7.11±0.12 <sup>c</sup>	4.36±0.43 <sup>b</sup>	0.75±1.22 <sup>a</sup>
Nickel	12.3±0.13 <sup>c</sup>	6.35±6.35 <sup>a</sup>	9.18±0.78 <sup>b</sup>
Lead	1.22±0.23 <sup>a</sup>	3.09±3.09 <sup>b</sup>	7.5±0.48 <sup>c</sup>
Zinc	29.74±0.23 <sup>ab</sup>	22.21±0.90 <sup>a</sup>	38.36±0.61 <sup>c</sup>
Iron	540±0.09 <sup>a</sup>	945±1.2 <sup>b</sup>	1074±0.13 <sup>c</sup>
Chromium	0.028±0.01 <sup>a</sup>	BDL	BDL
Cadmium	0.03±0.02 <sup>ab</sup>	BDL	0.01±0.01 <sup>a</sup>

Data are mean values±S.E in rows with different superscript alphabet are significantly different (p<0.05) DMRT.

## Discussion

It is very important to know about the chemical composition of the dominant seagrass species, because of their direct and indirect roles in coastal marine food chains. However, there are a few literatures that deal with the biochemical components in seagrass species.<sup>27-31</sup> Moreover, there is no recent data available on the biochemical components of the three seagrass species investigated, i.e., *Cymodocea serrulata*, *Halodule uninervis* and *Syringodium isoetifolium*. Dugong is a seagrass dependent marine mammal of tropical and subtropical coastal waters. It feeds mainly on seagrass species in the genera of *Zostera*, *Posidonia*, *Thalassia*, *Enhalus*, *Cymodocea*, *Halodule*, *Syringodium*, and *Halophila*.<sup>32</sup> It generally selects the fast growing species of *Halodule uninervis* and *Halophila ovalis* over the sympatric slow growing *Cymodocea rotundata*, *Thalassia hemprichii*, and *Zostera capricorni*.<sup>33</sup> Seagrasses such as *Enhalus acoroides*, *H. ovalis*, *H. spinulosa*, *Cymodocea serrulata*, *H. pinifolia*, *H. uninervis* and *Syringodium isoetifolium* are food for large species, such as *Dugong* (dugong, status: vulnerable, VU A1cd) and turtle *Chelonia mydas* (green turtle, status: endangered, EN A1bd). Approximately over half of seagrasses are known to be consumed by sea mammals in the Palk bay because many marine and estuarine organisms feed on seagrasses.<sup>30</sup> Heck and Valentine<sup>34</sup> found that sea urchin, *Lytechinus ariegates*, consumes 50 to 100% of the above ground seagrass biomass. Similarly, Keller<sup>35</sup> and Jernakoff et al.,<sup>36</sup> reported that *Tripneustes ventricosus* and *Diadema antillarum* consume large quantities of seagrass. Turtles choose their seagrass in situ based on toughness, succulence, texture or taste which might

reflect plant age and health. Marine herbivorous fish diet choices are based on digestibility of cell wall and storage of carbohydrates as well as nutrient and energy content.<sup>37</sup> Thus, sea grasses are of great nutritive value for all marine organisms that feed them, such as sea urchins, sea turtles, dugong, sea manatees.<sup>38</sup> Apart from that seagrasses are used as medicine to treat various skin diseases, burns and boils by the people of South India fishing communities of Tamil Nadu.<sup>39</sup> A unique marine based product can give a useful effect to the body, for example, anti-HIV agents isolated from *Thalassia testudinum* was beneficial from a medical perspective.<sup>40</sup> Seagrasses have a high calorific value that can provide their great economic values and their importance in the food chain. Thus, the study on the macro and micro nutrient content of seagrasses in Gulf of Mannar provides evidence of the health status of not only seagrasses but also the surrounding environments and supported marine ecosystem.

## Proteins

Normally protein content of seagrass is lower (12-19%) than that of the animal (23%). Hence, they cannot realistically be regarded as a significant source of dietary protein for marine animals. The dietary protein requirement of marine animals is met by some species of seaweed that contain high protein levels. Suberkropp et al.,<sup>41</sup> reported that seagrasses are mostly low in protein content. Augier et al.,<sup>16</sup> reported protein content is high in the whole seagrass plant compared to that of the leaves alone. Kannan<sup>42</sup> have reported that the maximum protein content of *E. acoroides* leaves is 7.11% but in rhizome it is 15.93%. In the present study, the whole plant was taken for the analysis, so that the nutritive composition was higher than the leaves

of seagrass (6%) already reported by Price.<sup>43</sup> The results suggested that the rhizomes and shoots are the storage organs from which soluble carbohydrates and proteins can be mobilized for plant growth.<sup>44</sup> The seasonal variation does not affect the protein content of seagrass and it was evident from the study of Pradheeba et al.,<sup>30</sup> who reported similar protein levels in summer and monsoon seasons.

### Lipid

Lipid content in the seagrass was not a striking feature as it showed only very little difference between the species. In the present study, all the seagrass species had high lipid content. Shams El Din & El-Sherif<sup>45</sup> reported that the lipid content in seagrasses is high in rhizomes than that in the leaves. Pirc<sup>46</sup> reported that variations in the total lipid content of the seagrasses could be attributed to the age, stage of growth and ecological variations.

### Carbohydrate

Higher carbohydrate content was found in *C. serrulata*. This is because, the leaves and rhizomes of seagrass act as the reservoirs for food storage. This variation is largely related to the profuse rhizome systems of the seagrass species. Species like *C. rotundata*, *C. serrulata*, *E. acoroides* and *T. hemprichii*, which are having thick rhizomes, registered higher carbohydrate content. In the present study also, *C. serrulata* showed higher carbohydrate than *H. univervis* and *Syringodium isoetifolium*. Kannan<sup>43</sup> recorded 21.14% of carbohydrate in *C. serrulata*, 8.52% carbohydrate in *H. univervis*. This indicates that there is a clear cut spatial variation in the biochemical content of seagrass species. The carbohydrate content of seagrasses also depends on the nutrients available in its ambient environment.

### Chlorophyll

Pigment (Chlorophyll) composition in seagrass is similar to that of any other angiosperms and includes chlorophyll a and b, which are directly involved in photosynthesis. Chlorophyll content of the seagrass species can be largely influenced by the availability of the light and morphology of the seagrass leaves. The present results indicate that there is no variation in chlorophyll concentration between the three seagrasses by registering the similar value. This is because water column was clearer and the light availability was the same for the seagrasses investigated. Moreover, there was no light stress, which is considered to be the important parameter for maintaining chlorophyll content and other photosynthetic growth parameters of seagrasses. The present study results coincide with that of Coles & Mckenzie,<sup>47</sup> who reported a high value of 57µg/g in *Ulva canopy* when the water column was clear with a higher quantum of irradiance. But Nelson et al.<sup>48</sup> reported that due to high irradiance intensity during the summer season, the chlorophyll content will be at its minimum of 30µg/g in *Ulva sp.* In addition to that, there was also a clear interspecies variation in chlorophyll concentration. The interspecies variation could be attributed to the morphology of the leaf and the depth in which the plants are growing. Seagrasses like *H. univervis* with lower leaf surface areas due to their linear leaf structure and *S. isoetifolium* with cylindrical leaves growing relatively in deeper waters, registered lower chlorophyll contents while species like *Cymodocea serrulata* with broad leaf width recorded higher chlorophyll content.<sup>49</sup> In the present study also higher chlorophyll content was observed in *Cymodocea serrulata* due to their with broad leaf width.

### Ash

Ash usually represents the inorganic part of the plant. This is because ashing destroys all the organic material present in the sample. The ash content of seagrasses in the present study varied from 18.2 to 28.7%. The percentage of ash content is highest in *Halodule uninervis* (28.7%). The chemical analysis of the three marine plants in the present study showed high ash content. The high ash content is a general feature of seagrass and these values are generally much higher than those of terrestrial green leaves (15%).<sup>50</sup> High ash content invariably indicates the presence of appreciable amounts of diverse mineral components. The amounts of ash vary with phylum, season, environmental, geographical and physiological variations.<sup>51</sup> The amount of ash in plant material varies considerably according to the part of the plant, age etc. The constituents of the ash vary with time and from organ to organ. Kar et al.,<sup>52</sup> detected 18.53% of organic content and 26.22% of inorganic content in *T. cordifolia* and this is similar to our results. The results of this study are close to the results reported by Black & Kenney<sup>53</sup> and Durako & Dawes,<sup>54</sup> i.e., 35% ash content in marine plants.

### Fiber

Plant sources of dietary fiber are often classified into the soluble or insoluble fiber. Plant foods contain both types of fiber in various degrees, according to the plant's characteristics. The advantages of fiber in seagrass are the production of healthy compounds during the fermentation of soluble fiber and insoluble fiber's ability to increase bulk, soften stool and shorten transit time through the intestinal tract. The disadvantage of high fiber diet is the potential for significant intestinal gas production and bloating. Seagrass is regarded as a potentially rich source of polysaccharide carbohydrates for ruminants.<sup>55</sup> In this study, the crude fiber content of the seagrass ranged from 27 to 37% in this study. This is higher than the levels found in terrestrial plants, and the fibers in seagrasses are rich in soluble fractions.<sup>56</sup> Holdt & Kraan<sup>57</sup> reported that the seagrass dietary fibers contain some valuable nutrients and substances. Hence, there has been a great deal of interest in seagrass meal, functional foods and nutraceuticals for human consumption. In addition, seagrass has other health benefits, such as, polysaccharides in seagrasses show anti-tumour and anti-herpetic bioactivity, anticoagulant and decrease low-density lipid cholesterol in rats, prevent obesity, large intestine cancer and diabetes and have antiviral activities.

### Minerals

In all the three seagrass species investigated, the average concentrations of sodium, calcium and magnesium were higher compared to that of phosphorus and potassium. On the other hand, the concentrations of these elements varied greatly in comparison with that of the previous studies reported in literature.<sup>58-62</sup> The variations in mineral contents were attributed to metabolic reactions, environmental conditions and seasonal variations and to the different requirements of the plant. Barko & Smart<sup>63</sup> reported that rooted aquatic plants could facilitate mineral uptake by roots from the sediments. The mineral content of seagrasses also depends on the minerals concentrations in the interstitial and overlying waters. Mineral content of sediment was high in Gulf of Mannar.<sup>39</sup> Masoud et al.,<sup>62</sup> stressed the importance of carbonate sediments as the main source of minerals. Fourqurean & Cai<sup>59</sup> suggested that mineral content in seagrasses is controlled by freshwater and marine inputs of these elements. Corals are calcium

carbonate compounds and the sediments also have rich carbonates and these give beneficial effects to seagrasses. Yamamuro & Chirapart,<sup>64</sup> studied seagrasses collected along the coast of Thailand, indicated that *H. ovalis* contained more ash and minerals compared to other seagrasses such as *E. acoroides*, *T. hemprichii* and *C. rotundata*. In the present study phosphorus, potassium and magnesium content were higher in *C. serrulata*, sodium content was high in *H. uninervis* whereas calcium content was high in *S. isoetifolium*. Yamamuro & Chirapart<sup>64</sup> reported calcium content of *H. ovalis* and *H. spinulosa* collected along the coast of Thailand were 576 and 466mg/100g and magnesium content were 974 and 3534.67mg/100g and this coincided with the present study. The seagrass species analysed in the present study contained higher calcium (600–690mg/100g) compared to that of the seaweed species such as *Gracilaria changii* at 110mg kg<sup>-1</sup>.<sup>65</sup> According to Heaney,<sup>66</sup> in human and animal, calcium is important for bones. It is not possible either to build or maintain a fully normal skeletal mass without an adequate intake of Calcium. The comparative analysis suggested that there is a distinct variation of seagrass species in terms of macronutrient. Many factors e.g., chemical, physical and biological environment could influence the nutrient content in seagrass species.

### Trace elements

Many literatures dealt with the trace elements, such as, cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), Nickel (Ni), lead (Pb) and zinc (Zn) in seagrasses across different parts of the world, In India,<sup>45,77</sup> in Mediterranean Sea<sup>68–78</sup> and in Egyptian waters.<sup>62,79,80</sup> This is because the seagrasses are considered as one of the most suitable biological indicators for water quality.

Zn and Cu are essential for the growth of seagrass. In general, their concentrations are low, i.e., <10 µg/g for copper and <30µg/g for Zn.<sup>81</sup> The concentration of Cu in Green Algae (*Caulerpa toxifolia*) from the Mediterranean coast in France is in the range of 2.0 to 5.6µg/g.<sup>32</sup> Red Algae from Greece contain 2.2 to 407µg/g.<sup>82</sup> Brown Algae (*Fucus vesiculosus*) from Greenland contain 1.3 to 3.3µg/g of Cu.<sup>83</sup> While Cu concentration in seagrass (*Posidonia oceanica*) from Northwest, Mediterranean varies from 7.9 to 22.0µg/g.<sup>84</sup> Cu concentrations, for three species from this study, were lower than that of the above species reported in literature, this may be due to age of seagrass species. According to Ledent et al.,<sup>85</sup> metal concentration shows a significant decrease with age, where metal concentration tended to be higher in young leaves than in adult leaves. This is because, in adult leaves, part of the mineral content has been re-translocated or leached.<sup>20,25</sup> Heavy metal accumulations within seagrass appear to be influenced more by the levels of biologically available metals within the surrounding water column, rather than by the sediment load, where the heavy metals are generally complexed or precipitated.

Pb and Ni are generally characterized as toxic elements for living organisms. The concentration of Pb and Ni is below the toxic limit in all the three species in this study. St-Cyr & Campbell<sup>86</sup> stressed on the important role of the root system in mineral uptake. Mohamed<sup>87</sup> referred the variation in metal contents in plants to the physico-chemical parameters such as temperature, pH, salinity, wave exposure and light, which are also considerable in the present study, where nickel element was positively influenced by water depth and phosphorus concentration. On the other hand, Geneid & Mourad<sup>80</sup> attributed these variations to biological conditions, which influence the bioavailability of trace metals and attributed inter-specific differences.

Zn, Fe, Cu are essential micronutrients for plants and can be toxic at higher concentrations than normal amounts requirement for growth.<sup>17</sup> Trace elements of Zn have negative health effects if it is consumed in high quantities. According to FAO,<sup>88</sup> the limits of Cu, Pb, Ni and Zn in the biota should not exceed 10, 6, 20 and 30µg/100g respectively. Accordingly, the concentrations of Cu, Pb, Ni and Zn were lower in all the three seagrass species of the present study than the prescribed maximum limits. This indicates that the trace metals incorporated in the diet of the herbivorous animals, which feed on these seagrasses, cannot be bioaccumulated in their bodies. Hence, seagrasses do not affect the health and the organisms in the marine food chain.<sup>72</sup> Moreover, the surrounding environmental conditions of the seagrass species should be controlled to avoid any heavy metal pollution effect.

There is no available guideline about the limit of Fe in biota. However, Oliveira et al.,<sup>89</sup> have recorded a high value of 1000µg/100g in edible seaweeds. In the present study the content of Fe was high, whereas the content of chromium and cadmium were lowest in all three species in this study. Similar trends in content were reported for other seagrasses by Denton et al.,<sup>90</sup> They have reported that the concentration of cadmium, chromium and iron found in the seagrass were <5, <3 and <183µg/100g respectively. The authors suggested that a high concentration of chromium, cadmium or iron in seagrass would influence the intestinal absorption in the dugong and would also affect the livers and kidneys of dugongs. Syarifah et al.,<sup>91</sup> reported that the chromium content in *Halodule pinifolia* root-rhizomes and leaves were 6 and 6.99µg/100g respectively, while the cadmium concentration in root-rhizomes was 8.4µg/100g and in leaves, it was 6.08µg/100g.

Metals sequestered by seagrasses may be passed through trophic links to higher-level consumers, including, dugongs (*Dugong dugong*) and turtles (*Chelonia mydas*), which are the dominant consumers of seagrasses in tropical ecosystems.<sup>92</sup> Seagrasses also have particular promise for the detection of specific factors that may influence both short and long-term changes in the nearshore aquatic ecosystems. Heavy metal accumulations within seagrass leaf tissue appear to be influenced more by the levels of biologically available metals within the surrounding water column, rather than by the sediment load, where the heavy metals are generally complexed or precipitated. The concentrations of cadmium in the whole tissues of marine plants generally fall in the range of 0.001–27µg/g dry weight. Another laboratory study suggested that the ambient concentrations of dissolved cadmium in seawater may be high enough in some contaminated marine environments to cause harm to marine organisms. A short exposure to high concentration of dissolved cadmium in the laboratory would lead to accumulation of high concentrations of labile cadmium in tissues, whereas chronic exposure to low concentrations of cadmium in water and food in the natural environment would lead to high tissues residues of cadmium, most of which is tightly bound in a nontoxic form to tissue macromolecules (metallothionein) or granules. Therefore, it is possible that the benthic communities in areas with cadmium-contaminated sediments are adversely affected by the cadmium in the sediments.<sup>93</sup> Cadmium content of seagrass above the range of 3.5–33µg/100g has an adverse effect on reproduction.<sup>93</sup> Based on the recent publications about the concentration of cadmium in the whole tissues of marine plants from throughout the world, which are summarized by Neff,<sup>93</sup> cadmium concentration in phytoplankton is in the range of 0.04 to 4.6µg/g dry wt, in macro algae it varies

from 0.1 to 29.8 µg/g dry wt and in seagrasses it ranges between 1.0 and 4.9 µg/100g dry wt. The concentration of cadmium in all the three species investigated was lower than that reported in the literature. The chromium concentration in *C. serrulata* was 0.028 µg/100g and below the detectable level in *Halodule uninervis* and *Syringodium isoetifolium*. Bioconcentration of heavy metals by the marine plant is dependent on the concentrations of dissolved metals and the ratio to phosphorus in the ambient medium.<sup>94</sup> Besides, the accumulation is more rapid in the presence of sunlight than in the dark, suggesting that heavy metal bioaccumulation is at least partly energy dependent.

## Conclusion

The leaves of the three seagrass species, namely, *Cymodocea serrulata*, *Halodule uninervis* and *Syringodium isoetifolium* are found to be a rich source of biochemical components, major and essential trace elements. Hence, the seagrasses species are of great nutritive value for the marine organisms and play an essential role in the marine food chain. Nutritional composition data provided in this study would help to understand the dynamics of the marine mammals. Seagrasses in this studied area are a good candidate for bioremediation of polluted sites and bio-filtration of aquaculture effluent. It also could be considered as a good food and energy source for human, animals, fish and marine mammals. It is a promising raw material for several industries, such as cosmetic and biofuel industries. The analysis suggested that the concentration of macro- and micro-nutrients vary with seagrass species. Moreover, many factors (e.g., chemical, physical and biological environment) could influence the nutrient content in seagrass species.

## Acknowledgements

The authors are thankful to Dr. J.K. Patterson Edward, Director, Suganthi Devadason Marine Research Institute, India for providing us the facilities to carry out the work.

## Conflicts of interest

Author declares that there is none of the conflicts.

## References

- Wright JP, Jones CG. The concept of organisms as ecosystem engineers ten years on: progress, limitations, and challenges. *Journal of Bioscience*. 2006;56(3):203–209.
- Hemminga M, Duarte CM. *Seagrass Ecology*. UK: Cambridge University Press; 2000. 298 p.
- Gnassia-Barelli M, Lemee R, Pesando D, et al., Heavy metal distribution in *Caulerpa taxi-fofia* from the North- Western Mediterranean. *Mar Pollut Bull*. 1995;30(11):749–755.
- Haznedaroglu MZ, Zeybeck U. HPLC determination of chicoric acid in leaves of *Posidonia oceanica*. *Pharmaceutical Biology*. 2007;45(10):745–748.
- Aketa K, Kawamura A. Digestive functions in Sirenians and others (Review). In: *The Bulletin of Faculty of Biosources*. Mie University. 2001;27:85–103.
- Wood JR, Wood FE. Reproductive biology of captive green sea turtles *Chelonia mydas*. *Am Zool*. 1980;20(3):499–505.
- Abbott IA, Huisman JM. The marine green and brown algae of the Hawaiian Islands. Honolulu: Bishop Museum Press; 2004. 259 p.
- Abdel Hady HH, Daboor SM, Ghonemy AE. Nutritive and antimicrobial profiles of some seagrasses from Bardawil Lake, Egypt. *Egyptian Journal of Aquatic Research*. 2007;33(3):103–110.
- Papenbrock J. Highlights in seagrasses phylogeny, physiology, and metabolism: what makes them special? *Int Res Network*. 2012.
- Kannan L, Thangaradjou T. Identification and assessment of biomass and productivity of seagrass. National Training Workshop on Marine and coastal biodiversity assessment for conservation and sustainable utilization. *SDMRI Publication*. 2006;10:9–15.
- Kanna L. Seagrass of India. Ecobiology and conservation. In: *National symposium of marine plants, Their Chemistry and Utilization*. 2005.
- Ferrat L, Pergent-Martini C, Romeo M. Assessment of the use of biomarkers in aquatic plants for the evaluation of environmental quality: application to seagrasses. *Aquat Toxicol*. 2003;65(2):187–204.
- Nguyen VT, Ueng JP, Tsai GJ, et al. Proximate composition, total phenolic content, and antioxidant activity of Seagrape (*Caulerpa lentillifera*). *J Food Sci*. 2011;76(7):C950–C958.
- Caccia VG, Millero FJ, Palanques A. The distribution of trace metals in Florida Bay sediments. *Mar. Pollut. Bull*. 2003;46(11):1420–1433.
- Malea P, Haritonidis S, Kevrekidis T, et al. Seasonal and local variations of metal concentrations in the seagrass *Posidonia oceanica* (L.) Delile in the Antikyra Gulf, Greece. *Science of the Total Environment*. 1994;153(3):225–235.
- Augier H, Calvert H, Wollaston E, et al. A comparison of the C, H, N, protein and amino acid composition of *Posidonia australis* Hook. F. with that of *Posidonia oceanica* (L.) Delile and several other marine phanerogams. *Aquat Bot*. 1982;12:69–80.
- Kannan RRR, Arumugam R, Anantharaman P, et al. Chemometric studies of multielemental composition of few seagrasses from Gulf of Mannar, India. *Biol Trace Elem Res*. 2011;143(2):1149–1158.
- Kilminster K. Trace element content of seagrasses in the Leschenault Estuary, Western Australia. *Marine Pollution Bulletin*. 2013;73:381–388.
- Thangaradjou T, Nobil EB, Dilipan E, et al. Heavy metal enrichment in seagrass of Andaman Islands and its implication to the health of the coastal ecosystem. *Indian Journal of Marine Sciences*. 2010;39(1):85–91.
- Lowry OH, Rosebrough NJ, Farr AL, et al. Protein measurement with Folin phenol reagent. *Journal of Biological Chemistry*. 1951;193:265–275.
- Folch J, Less M, Stanley GHH. A simple method for the isolation and purification of total lipids from animal tissues. *J Biol Chem*. 1957;226(1):497–509.
- Dubois M, Gilles KA, Hamilton JK, et al. Colorimetric method of determination of sugars and related substances. *Analytical Chemistry*. 1956;28(3):350–356.
- Maynard AJ. *Methods in Food Analysis*. New York: Academic Press; 1970. 176 p.
- AOAC. Official methods of Analysis of AOAC International. 16th ed. USA: 1995.
- Lichtenthaler HK. Chlorophylls and carotenoids: pigments of photosynthetic. Biomembrane. *Methods Enzymol*. 1987;147:350–382.
- Birch WP. Some chemical and calorific properties of tropical marine angiosperms. *J Appl Ecol*. 1975;12(1):201–221.
- Dawes CJ, Bird K, Durako M, et al. Chemical fluctuations due to seasonal and cropping effects on an algal-seagrass community. *Journal of Aquatic Botany*. 1979;6:79–86.

28. Geneid YA, El-Hady HH. Distribution, biomass and biochemical contents of the seagrasses species of Lake Bardawil. *Biology and Marine Mediterranean*. 2006;13(4):225–229.
29. Mascaro O, Silvia Oliva S, Perez M, et al. Spatial variability in ecological attributes of the seagrass *Cymodocea nodosa*. *Botanica Marina*. 2009;52:429–438.
30. Pradheeba M, Dilipan E, Nobi EP, et al. Evaluation of seagrasses for their nutritional value. *Indian Journal of Geo-Marine Science*. 2011;40(1):105–111.
31. Tobatinejad NM, Annison G, Rutherford–Markwick K, et al. Structural constituents of the seagrass *Posidonia australis*. *Journal of Agricultural Food Chemistry*. 2007;55(10):4021–4026.
32. McRoy CP, Helfferich C. Applied aspects of seagrasses. In: *Handbook of Seagrass Biology: An Ecosystem Perspective*. Phillips RC, Mc Roy CP, editors. New York: Garland STPM Press; 1980. p. 297–343.
33. Preen A. Impacts of dugong foraging on seagrass habitats: observational and experimental evidence for cultivation grazing. *Mar Ecol Prog Ser*. 1995;124:201–213.
34. Heck KL, Valentine JF. Sea urchin herbivory: evidence for long-lasting effects in subtropical seagrass meadows. *Journal of Experimental Marine Biology and Ecology*. 1995;189(1–2):205–217.
35. Keller BD. Coexistence of sea urchins in seagrass meadows: an experimental analysis of competition and predation. *Journal of Ecology*. 1983;64:1581–1598.
36. Jernakoff P, Brearley A, Nielsen J. Factors affecting grazer–epiphyte interactions in temperate seagrass meadows. In: Ansell AD, Gibson RN, Barnes M, editors *Oceanography and Marine Biology: An Annual Review*. London: UCL Press. 1996;34:109–162.
37. Zemke–White WL, Clements KD. Chlorophyte and rhodophyte starches as factors in diet choice by marine herbivorous fish. *J Exp Mar Biol Ecol*. 1999;240:137–149.
38. Nagelkerken I, Dorenbosch M, Verberk WCEP, et al. Importance of shallow water biotopes of a Caribbean bay for juvenile coral reef fishes: patterns in biotope association, community structure and spatial distribution. *Marine Ecology Progress Series*. 2000;202:175–192.
39. Kannan L, Thangaradjou T, Anantharaman P. Status of seagrass of India. *Seaweed Res Utiln*. 1999;21:25–33.
40. Rowley DC, Hansen MST, Rhodes D, et al. Thalassiolins A–C: new marine–derived inhibitors of HIV cDNA integrase. *Bioorganic and Medicinal Chemistry*. 2002;10(11):3619–3625.
41. Suberkropp KF, Godshalk GL, Klung MJ. Changes in the Chemical Composition of Leaves During Processing in a Woodland Stream. *Ecology*. 1976;57(4):720–727.
42. Kannan R, Kannan L. *Biochemical and caloric composition of seaweeds and seagrass of the Palk Bay, South East coast of India*. In: VN Raja Rao editor. *Algological research in India: Festschrift*. In: Anand Bishen Singh N, Mahendra Pal Singh, Dehra Dun, editors. 2002. 471 p.
43. Pirc H. Growth Dynamics in *Posidonia oceanica*. I: Seasonal Changes of Soluble Carbohydrates, Starch, Free Amino Acids, Nitrogen and Organic Anions in Different Parts of Plant. *Mar Ecol*. 1985;6:141–165.
44. Dawes CJ, Lawrence JM. Seasonal changes in the proximate constituents of the seagrasses *Thalassia testudinum*, *Halodule wrightii* and *Syringodium filiforme*. *Journal of Aquatic Botany*. 1980;8:371–380.
45. Shams El Din NG, El–Sherif ZM. Nutritional value of *Cymodocea nodosa* and *Posidonia oceanica* along the western Egyptian Mediterranean coast. *Egyptian journal of aquatic research*. 2013;39(3):153–165.
46. Pirc H. Seasonal changes in soluble carbohydrates, starch and energy content in Mediterranean seagrasses. *Journal of Marine Ecology*. 1989;10(2):97–106.
47. Coles R, Mc Kenzie L. Trigger points and achieving targets for managers. Townsville: 2004.
48. Nelson TA, Haberlin K, Nelson AV, et al. Ecological and physiological controls of species composition in green macroalgal blooms. *Ecology*. 2008;89(5):1287–1298.
49. Drew EA. Sugars, cyclitols and seagrass phylogeny. *Aquat Bot*. 1983;15(4):387–408.
50. Ruperez P. Mineral Content of Edible Marine Seaweeds. *Food Chemistry*. 2002;79(1):23–26.
51. Mendis E, Kim SK. Present and future prospects of seaweeds in developing functional foods. In: *Advances in food and nutrition research*. Volume 64: Marine medicinal foods; Implications and applications, Macro and Microalgae. USA: Elsevier Inc; 2011. p. 1–13.
52. Kar A, Choudhary BK, Bandyopadhyay NG, et al. Preliminary studies on the inorganic constituents of some indigenous hypoglycaemic herbs on oral glucose tolerance test. *J Ethnopharmacol*. 1999;64:179–184.
53. Black JL, Kenney PA. Factors Affecting Diet Selection by Sheep. II. Height and Density of Pasture. *Australian Agric Res*. 1984;5:565.
54. Durako MJ, Dawes CJ. A comparative seasonal study of two populations of *Hypnea musciformis* from the east and west coasts of Florida, USA. I. Growth and Chemistry. *Mar Biol*. 1980;59(3):151–156.
55. Kailas A, Nair S. Comparison of nutrient compositions and calorific values of eight tropical seaweeds. *Phykos*. 2015;45(1):62–74.
56. Dawczynski CH, Schafer U, Leiterer M, et al. Nutritional and toxicological importance of macro, trace, and ultra-trace elements in algae food products. *J Agric Food Chem*. 2007;55(25):10470–10475.
57. Holdt SL, Kraan S. Bioactive compounds in Seaweed: Functional food applications and legislation. *Journal of Applied Phycology*. 2011;23(3):543–597.
58. Delgado O, Ruiz J, Perez M, et al. Effects of fish farming on seagrass (*Posidonia oceanica*) in a Mediterranean bay: seagrass decline after organic loading cessation. *Oceanologica Acta*. 1999;22(1):109–117.
59. Fourqurean JW, Cai Y. Arsenic and phosphorus in seagrass leaves from the Gulf of Mexico. *Aquatic Botany*. 2001;71(4):247–258.
60. Kannan R, Ganesan M, Govindasamy C, et al. Tissue concentration of Heavy metals in seagrasses of the Palk Bay, Bay of Bengal. *Int J Eco Env Sc*. 1992;18:29–34.
61. Kannan RR, Arumugam R, Anantharaman P. Chemometric studies of multielemental composition of five seagrasses from Gulf of Mannar, India. *Journal of Biological Trace Element Research*. 2010;143(2):1149–1158.
62. Masoud MS, El–Sarraf WM, Harfoush AA, et al. The effect of fluoride and other ions on algae and fish of coastal water of Mediterranean Sea, Egypt. *American Journal of Environmental Science*. 2006;2:49–59.
63. Barko JW, Smart RM. Mobilization of sediment phosphorus by submersed freshwater macrophytes. *Freshwater Biology*. 1980;10(3):229–239.
64. Yamamuro M, Chirapart A. Quality of the seagrass *Halophila ovalis* on a Thai intertidal flat as food for the Dugong. *Journal of Oceanography*. 2004;61(1):183–186.
65. Norziah MH, Ching CY. Nutritional composition of edible seaweed *Gracilaria changgi*. *Food Chemistry*. 2000;68(1):69–76.
66. Heaney RP. Bone mass, nutrition and other lifestyle factors. *Nutrition Reviews*. 1996;54(4 Pt 2):S3–S10.

67. Riosmena-Rodríguez R, Talavera-Saenz A, Costa-Vargas B, et al. Heavy metals dynamics in seaweeds and seagrasses in Bahía Magdalena, B.C.S., Mexico. *Journal of Applied Phycology*. 2010;22:283–291.
68. Campanella L, Conti ME, Cubadda F, et al. Trace metals in seagrass, algae and mollusks from an uncontaminated area in the Mediterranean. *Journal of Environmental Pollution*. 2001;111(1):117–126.
69. Conti ME, Bocca B, Iacobucci M, et al. Baseline trace metals in seagrass, algae, and mollusks in a Southern Tyrrhenian ecosystem (Linosa Island, Sicily). *Archives of Environmental Contamination and Toxicology*. 2010;58(1):79–95.
70. Conti ME, Iacobucci M, Cecchetti G. A biomonitoring study: trace metals in seagrass, algae and mollusks in a marine reference ecosystem (Southern Tyrrhenian Sea). *International Journal of Environmental Pollution*. 2007;29:308–332.
71. Dileo A, Annicchiarico C, Cardellicchio N, et al. Trace metal distributions in *Posidonia oceanica* and sediments from Taranto Gulf (Ionian Sea, Southern Italy). *Journal of Mediterranean Marine Science*. 2013;14(1):204–213.
72. Gardner SC, Fitzgerald SL, Acosta Vargas B, et al. Heavy metal accumulation in four species of sea turtles from the Baja California Peninsula, Mexico. *Biomonitoring*. 2006;19(1):91–99.
73. Garcia SMP, Olive I, Brun FG, et al. Non-structural carbohydrates and elemental composition in Seagrasses: an indicator of seagrass meadow health. In: *Mediterranean Seagrass Conference*. Malta: 2006.
74. Pergent-Martini C, Buia MC, Kantin R, et al. Evaluation of metal contamination based on *Posidonia oceanica*. In: Ozhan E, editor. *Proceeding of the Eighth International Conference of the Mediterranean Coastal Environment*. 2007;807–817.
75. Richir J, Gobert S, Sartoretto S, et al. 2010. *Posidonia oceanica* (L.), Delile, a useful tool for the biomonitoring of chemical contamination along the Mediterranean coast: A multiple trace element study. In: El Asmi S, Langar H, Belgacem W, editors. *Proceedings of the Fourth Mediterranean Symposium on Marine Vegetation*. 2010. 251 p.
76. Sanz-Lázaro C, Malea P, Apostolaki ET, et al. The role of the seagrass *Posidonia oceanica* in the cycling of trace elements. *Journal of Biogeosciences Discussion*. 2012;9:2623–2653.
77. Sawidis T, Brown MT, Zachariadis G, et al. Trace metal concentrations in marine macroalgae from biotopes in the Aegean Sea. *Environment International*. 2001;27(1):43–47.
78. Tranchina L, Brai M, Dagostino F, et al. Trace metals in *Posidonia oceanica* seagrass from South– Eastern Sicily. *Chemistry and Ecology*. 2005;21(2):109–118.
79. El-Deeb KM, Aboul-Naga WF. Trace metals: Fe, Zn, Mn, Cu, Ni, and Cr in macroalgae from Alexandria coast. *Bulletin of Faculty of Science*. Alexandria University. 2002;42(1,2):51–60.
80. Geneid YA, Mourad F. Levels of trace metals in the seagrasses of Lake Bardawil (Eastern Mediterranean, Egypt). *Actes du 3eme Symposium mediterraneen sur la vegetation marine*. 2007;(27–29):62–69.
81. Round FE. *The Biology of the Algae*. London: Edward Arnold Ltd; 1973. 278 p.
82. Malea P, Haritonidis S. Local distribution and seasonal variation of Fe, Pb, Zn, Cu, Cd, Na, K, Ca and Mg concentrations in the seagrass *Cymodocea nodosa* (Ucria) Aschers. in the Antirya Gulf, Greece. *Mar Ecol*. 1995;16(1):41–56.
83. Riget F, Johansen P, Asmund G. Natural seasonal variation of cadmium, copper, lead and zinc in brown seaweed (*Fucus vesiculosus*). *Mar Pollut Bull*. 1995;30(6):409–413.
84. Warnau M, Kedent G, Temura A, et al. Allometry of heavy metal bioconcentrations in the echinoid *Paracentrotus lividus*. *Arch Environ Contam Toxicol*. 1995;66:187–195.
85. Ledent G, Mateo MA, Warnau M, et al. Element losses following distilled water rinsing of leaves of the seagrass *Posidonia oceanica* (L.) Delile. *Aquatic Botany*. 1995;11:229–235.
86. St-Cyr L, Campbell PGC. Bioavailability of sediment-bound metals for *Vallisneria americana* Michx, a submerged aquatic plant, in the St. Lawrence River. *Canadian Journal of Fisheries and Aquatic Sciences*. 2000;57(7):13–30.
87. Mohamed HAE. Heavy metals in Suez Canal relevant to the impacts of land based sources. Ph.D. Thesis, University of Mansoura, Faculty of Science, Department of Chemistry. 2002. 223 p.
88. Food and Agriculture Organization, FAO. Committee for inland fisheries of Africa. Report of the third session of the working party on pollution and fisheries, FAO Fisheries report No. 471. 1992.
89. Oliveira MN, Freitas AL, Carvalho AF, et al. Nutritive and non-nutritive attributes of washed-up seaweeds from the coast of Ceara, Brazil. *Food Chemistry*. 2009;115(1):254–259.
90. Denton GRW, Marsh H, Heinsohn GE, et al. The unusual metal status of the *Dugong dugon*. *Mar. Pollut Bull*. 1980;4(9):135–138.
91. Syarifah NTB, Noor AMS, Sitiashhah A, et al. Experimental and field study on accumulation of heavy metals in seagrass (*Halodule pinifolia* and *Halophila minor*) in setiu wetland, Terengganu. *Journal of sustainability science and management*. 2008;3(1):41–73.
92. Lanyon JM, Limpus CJ, Marsh H. Dugongs and turtles: grazers in the seagrass system. In: Marsh H, Larkum AWD, Mc Comb AJ, Shepherd SA, editors. *Biology of seagrasses. A treatise on the biology of seagrasses with special reference to the Australian region*. Aquatic Plant Studies 2. Amsterdam: Elsevier Science Publishers BV; 1989. p. 610–634.
93. Neff JM. Bioaccumulation in marine organism. Effect of contaminants from oil well produced water. Elsevier. 2002;145–151.
94. Kudo I, Kokubun H, Matsunaga K. Chemical fractionation of phosphorus and cadmium in the marine diatom *Phaeodactylum tricorutum*. *Mar Chem*. 1996;52:221–231.