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Evaluation of seagrasses for their nutritional value

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Biochemical and calorific contents of different species of seagrasses occurring in Palk Bay were determined: *Enhalus acoroides* (68.82 K cal g^{-1}), *Halophila beccarii* (29.16 K cal g^{-1}), *Halophila ovalis* (24.08 K cal g^{-1}), *Cymodocea rotundata* (60.62 K cal g^{-1}), *Cymodocea serrulata* (43.40 K cal g^{-1}), *Halodule uninervis* (40.15 K cal g^{-1}), *Halodule pinifolia* (28.39 K cal g^{-1}) and *Syringodium isoetifolium* (38.38 K cal g^{-1}) of leaves and in rhizome *Enhalus acoroides* (77.84 K cal g^{-1}), *Halophila beccarii* (38.84 K cal g^{-1}), *Halophila ovalis* (37.03 K cal g^{-1}), *Cymodocea rotundata* (63.68 K cal g^{-1}), *Cymodocea serrulata* (37.33 K cal g^{-1}), *Halodule uninervis* (28.39 K cal g^{-1}), *Halodule pinifolia* (28.23 K cal g^{-1}) and *Syringodium isoetifolium* (30.48 K cal g^{-1}). Calorific contents of seagrasses were equivalent to Bengal gram, Peas, Potato and Sweet potatoes. Present study suggests that the seagrasses could be considered as feed/food.

[**Key Words**: Seagrass, chlorophyll, debris, shore]

Introduction

Seagrasses can be found distributed from the mid-intertidal areas to depths greater than 50 m[1]. In India, 14 species of seagrasses have been recorded along the east and west coasts[2,3]. The Gulf of Mannar, Palk Bay, Andaman and Nicobar islands and Lakshadweep islands are known for their seagrass resources. Seagrass species *viz. Enhalus acoroides, Thalassia hemprichii, Cyomodocea* spp. and *Halodule* spp. contribute more biomass in these regions and their photosynthetic productivity is also higher as compared to the other most productive ecosystems of the world[4]. The fact that this abundant source is subjected to relatively low levels of direct grazing on the living plant material has prompted studies on the chemical constituents and relative food value of seagrasses[5]. Though 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 of the dominant seagrass species of the Palk Bay and to work out the calorific content so as to evaluate them for use as food for human consumption.

Materials and Methods

Seagrass samples were collected from the Palk Bay, Bay of Bengal during April 2007-March 2008 (April to June: Summer season; July to September: Pre-Monsoon season; October to December: Monsoon season; January to March: Post Monsoon season) at Devipattinum. The Palk Bay is a shallow basin located along the southeast coast with an average depth of 9 m mainly with muddy bottom at the shore regions and it covers an area of about 600 km^2[6]. This type of shallow coast with muddy bottom usually supports luxuriant growth of seagrasses. The predominant seagrasses occurring in this area are *Enhalus acoroides*, *Halophila ovalis*, *H. beccarii* and *Cymodocea serrulata*.

For the present study, 8 species of seagrasses (*E. acoroides, H. beccarii, H. ovalis, C. rotundata, C. serrulata, H. uninervis, H. pinifolia* and *S. isoetifolium*) were collected and analyzed for pigment and biochemical constituents. Seagrass samples were collected from the field by hand-picking method and washed thoroughly by using seawater to remove epiphytes, sand and debris and then transported to the laboratory. Leaves and rhizomes of individual species were separated and the fresh leaves were taken for chlorophyll estimation and the rest of them were dried for 7 days in shade. After proper drying the samples were estimated for, biochemical composition viz. carbohydrate, protein, lipid and secondary metabolites like phenol and tannin of leaves and rhizome using standard methods.

Concentration of chlorophyll a, b and total chlorophyll was determined spectrophotometrically[7] in 90% acetone extracts of fresh seagrass leaves. Dried seagrass sample was taken for carbohydrate estimation by following anthrone method[8] using glucose as standard. Protein was estimated by following the method described by Lowry *et al.*[9] using Bovine Serum Albumin as standard. Lipid was determined by means of gravimetric method using chloroform–methanol extraction[10]. Phenol was estimated, following the method described by Hendry[11] involving the procedures of Forrest and Bendall[12] by using different concentrations of catechol as standard and tannin was determined, following the method of Mole and Waterman[13].

Differences between biochemical constituents were studied using ANOVA with repeated measures (biochemical concentration), two-fixed factor (seagrass leaf and seagrass rhizome factor) and season factor. One way ANOVA was used to confirm the heterogeneity of the parameters studied at the different seasons. Statistical analysis was carried out using a statistical package (SPSS 11.5 for Windows) and $p<0.05$ was considered statistically significant.

All organic substances have a characteristic combustion heat. This makes it relatively easy to calculate the calorific content of an organism once the chemical constituents of its organic fraction have been determined by using standard equivalents. In the present study, the following known conversion values [14] were used to convert the organic content into calorific values: fats 9.45, carbohydrates 4.10 and protein 5.65 kcal g^{-1}.

Results

Total chlorophyll content of the test seagrasses varied from 0.05 to 0.29 mg g^{-1} while the chlorophyll a and b values ranged from 0.02 to 0.19 mg g^{-1} and 0.03 to 0.149 mg g^{-1} respectively (Figs. 1, 2 & 3). In general, all the species recorded lower total chlorophyll and chlorophyll a and b contents during the monsoon season and the higher contents, during the summer season. Significant positive correlation was obtained between different fractions of chlorophyll of the seagrass species ($p<0.001$). ANOVA indicated the significant variation in the concentration of chlorophyll-a and total chlorophyll content in different seagrass species ($F=11.807$, $p<0.001$).

Carbohydrate content of the seagrass leaves and rhizomes varied from 2 to 8.7 mg g^{-1} and 3 to 9.1 mg g^{-1} respectively (Figs. 4 & 5). Among all the species studied, *H. beccarii* recorded lower carbohydrate content in the leaves whereas *C. rotundata* recorded higher content both in the leaves and rhizomes. Monsoon season showed very low values of carbohydrate content when compared to other seasons in all the species. Carbohydrate content of the seagrass leaves showed significant positive correlation with protein content of leaves ($r=0.007$, $p<0.01$) and lipid in rhizome ($p<0.01$).

Protein content of seagrass leaves varied between 0.1 and 5.9 mg g^{-1} with a minimum of 0.1 mg g^{-1} in *H. beccarii* and a maximum of 5.9 mg g^{-1} in *E. acoroides* (Fig. 6). The protein content in rhizomes registered the minimum (0.6 mg g^{-1}) during the monsoon season in *T. hemprichii* while the maximum (7.2 mg g^{-1}) was recorded during the summer season in *E. acoroides* (Fig. 7). In general, monsoon season recorded lower protein value when compared to other seasons in all the species. Protein content showed

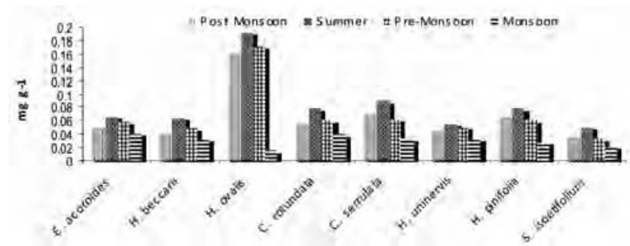

Fig. 1—Chlorophyll-a content in different species of seagrasses.

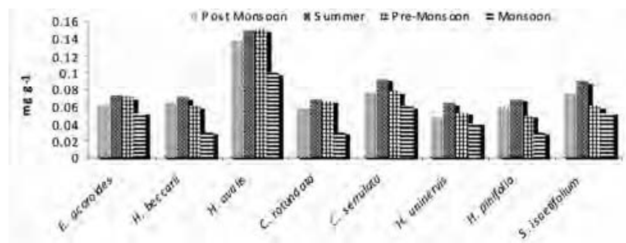

Fig. 2—Chlorophyll-b content in different species of seagrasses.

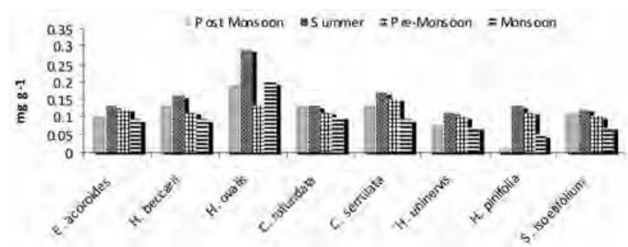

Fig. 3—Total Chlorophyll content in different species of seagrasses.

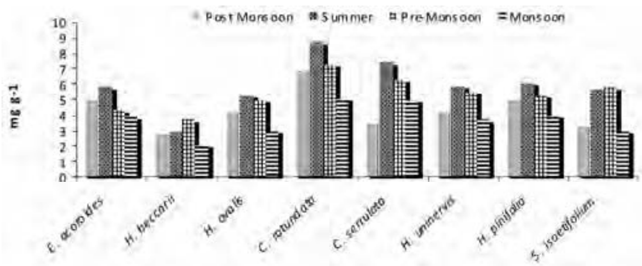

Fig. 4—Carbohydrate content recorded in the leaves of different species of seagrasses.

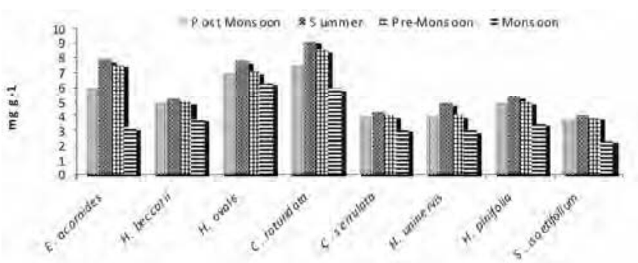

Fig. 5—Carbohydrate content recorded in the rhizomes of different species of seagrasses

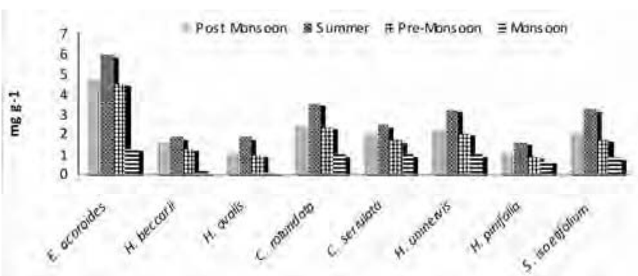

Fig. 6—Protein content recorded in the leaves of different species of seagrasses.

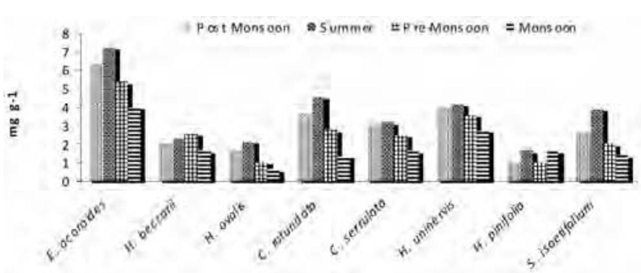

Fig. 7—Protein content recorded in the rhizomes of different species of seagrasses.

significant positive correlation between lipid, phenol and tannin of seagrass leaves and rhizomes (r=0.006, p<0.01), whereas significant variation was found in leaves (F=3.906) and rhizomes (F=9.935) of seagrass species with respect to monsoon season.

Lipid content of seagrass leaves and rhizomes varied from 0.01 to 3.2% and 0.03 to 4.1% respectively (Figs. 8 & 9). Monsoon season recorded very low lipid values when compared to other seasons in almost all the species except *H. uninervis*. Lipid content correlated positively with phenol and tannin (p<0.01) of seagrass leaves and rhizomes, whereas significant variation was observed in leaves (F=10.098) and rhizomes (F=16.384) in respect of seasons.

Phenol content of seagrass leaves varied between 3.2 and 9.2 mg g^{-1}, whereas, in rhizomes, phenol content ranged from 0.91 to 8.6 mg g^{-1}. Both leaf and rhizomes recorded the minimum values in *H. beccarii* and the maximum values in *E. acoroides* (Figs. 10 & 11). Significant positive correlation was found between phenol and tannin contents of seagrass leaves and rhizomes (p<0.05).

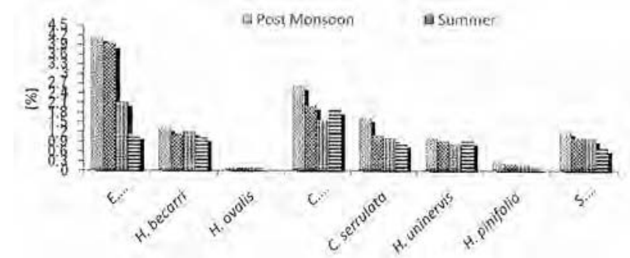

Fig. 8—Lipid content recorded in the leaves of different species of seagrasses.

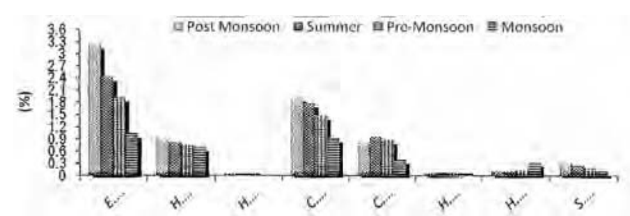

Fig. 9—Lipid recorded content in the rhizome of different species of seagrasses.

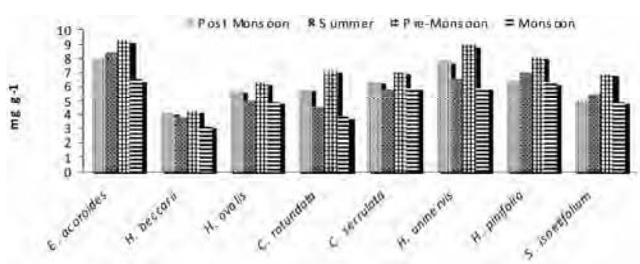

Fig. 10—Phenol content recorded in the leaves of different species of seagrasses.

Tannin content of seagrass leaves and rhizomes varied from 0.56 to 2.4 mg g^{-1} and 0.56 to 1.9 mg g^{-1} respectively (Figs. 12 & 13). In leaves, the minimum of 0.56 mg g^{-1} was recorded in *H. uninervis* during the summer season and the maximum of 2.4 mg g^{-1} was recorded in *E. acoroides* during the pre-monsoon season. Whereas, in rhizomes, the tannin content ranged from 0.56 to 1.9 mg g^{-1} registering the minimum in *H. pinifolia* during the summer season and the maximum in *E. acoroides* during the postmonsoon season. Tannin content showed much significant variations in leaves (F=2.629) and rhizomes (F=4.832) of seagrass species, with respect to seasons.

Calorific values of the seagrasses were calculated from biochemical constituents by using the universally accepted standard conversion factors. Calorific values were estimated individually for each

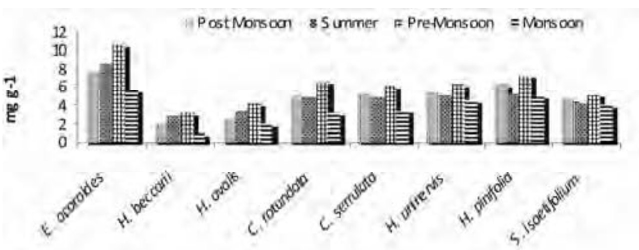

Fig. 11—Phenol content recorded in the rhizomes of different species of seagrasses.

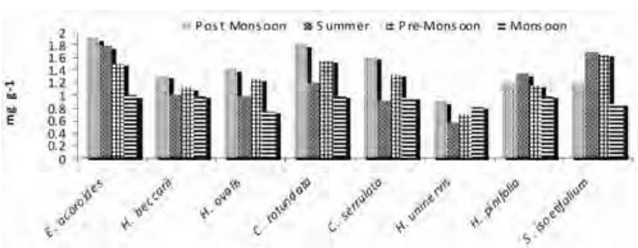

Fig. 12—Tannin content recorded in the leaves of different species of seagrasses.

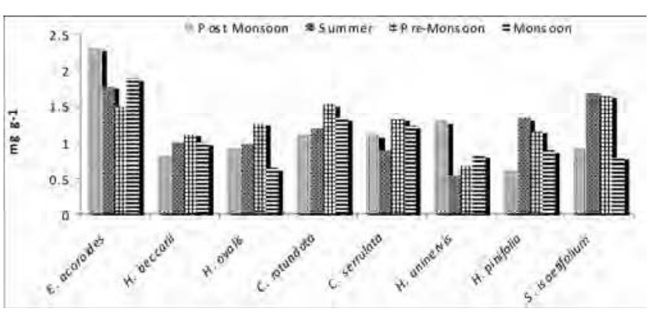

Fig. 13—Tannin content recorded in the rhizomes of different species of seagrasses.

constituent and summed for total calorific value of the species. Among all the seagrasses species studied, *E. acoroides* recorded maximum calorific content (68.82 k cal g^{-1}) followed by *C. rotundata* (60.62 k cal g^{-1}) and *C. serrulata* (43.40 k cal g^{-1}). *T. hemprichii* recorded lower calorific content (24.08 k cal g^{-1}) in leaves. Whereas rhizomes of *E. acoroides* (77.84 k cal g^{-1}) and *C. rotundata* (63.68 k cal g^{-1}) recorded higher calorific contents and *H. pinifolia* recorded minimum calorific value (28.23 k cal g^{-1}) (Table 1).

Discussion

Present study was carried out on the pigments (chlorophyll a, b and total chlorophyll), biochemical composition (carbohydrate, protein and lipids) and secondary metabolites (phenols and tannins) of the seagrass species viz. *E. acoroides*, *H. beccarii*, *C. rotundata, C. serrulata, T. hemprichii*, *H. uninervis*, *H. pinifolia* and *S. isoetifolium* and the calorific content of the individual seagrass species was calculated from the biochemical constituents.

Pigment composition in seagrasses is similar to that of any other angiosperms by including chlorophyll-a and b, which are directly involved in photosynthesis[15,16]. Chlorophyll content of the seagrass species can be largely influenced by the availability of the light and morphology of the seagrass leaves. Present results indicated that there is a clear seasonal variation in chlorophyll concentration by registering the minimum value during the monsoon season when the water column was more turbid and

Table 1—Calorific content in leaves and rhizomes (k Cal g^{-1}) of the seagrasses

Seagrass	Plant parts	Calorific content (k Cal g^{-1})
E. acoroides	Leaves	68.82
	Rhizomes	77.84
H. beccarii	Leaves	29.16
	Rhizomes	38.84
H. ovalis	Leaves	24.08
	Rhizomes	37.03
C. rotundata	Leaves	60.62
	Rhizomes	63.68
C. serrulata	Leaves	43.40
	Rhizomes	37.33
H. uninervis	Leaves	40.15
	Rhizomes	37.16
H. pinifolia	Leaves	28.39
	Rhizomes	28.23
S. isoetifolium	Leaves	38.38
	Rhizomes	30.48

the light availability to the seagrasses was limited causing light stress which is considered to be the important parameter for decreasing chlorophyll-a content and other photosynthetic growth parameters of seagrasses[17]. Chlorophyll recorded higher values during the summer season when the water column was clear with higher quantum of irradiance. In addition to the seasonal variations, there was also a clear inter-species variation in chlorophyll concentration. This inter-species variation could be attributed to the morphology of the leaf and the depth in which the plants are growing. Seagrass like *H. uninervis* and *H. pinifolia* 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 *H. ovalis* with broad leaf width recorded higher chlorophyll content[18].

There was a distinct seasonal variation in the carbohydrate content, both in the leaves and rhizomes, the later recorded comparatively higher carbohydrate content than the leaves. This is quite natural that the rhizomes are largely acting as the reservoirs for food storage. It is also found marked inter-species variations in carbohydrate content, largely relating to the profuse rhizome systems of the seagrass species. Two-way ANOVA revealed that there was more significant variation in carbohydrate content of leaves (F=3.734) and rhizome (F=8.54). Species like *C. rotundata*, *C. serrulata*, *E. acoroides* and *T. hemprichii* which are having thick rhizomes registered higher carbohydrate content.

The dominant storage carbohydrate in most seagrasses is the soluble product of sucrose and the other soluble carbohydrate includes glucose and fructose. The seasonal changes[19-22] of sucrose could be considerable with a minimum in the monsoon and the maximum in summer season[23]. Kannan and Kannan[24] recorded 1.14 mg g^{-1} of carbohydrate in *E. acoroides* and 1.52 mg g^{-1} in *T. hemprichii* in Kattumavadi and Kottaipatinam areas. However, in the present study, more amount of carbohydrate was recorded in leaf (8.7 mg g^{-1}) and rhizome (7.5 mg g^{-1}) of *C. rotundata*. This indicates that there is also clear cut spatial variation in the biochemical content of seagrass species, but this variation would depend on the nutrients available in the ambient environment.

Like carbohydrate, protein also recorded higher concentrations in rhizome when compared to the leaves. Higher concentration of protein was recorded in *E. acoroides* rhizome (7.2 mg g^{-1}) than the leaves (5.9 mg g^{-1}). Kannan and Kannan[24] have reported that the maximum protein content in *E. acoroides* was 15.93 mg g^{-1}. Interestingly, both the studies have reported higher protein content during the summer season. Like carbohydrate, protein concentration was also higher in *E. acoroides*, *Cymodocea* spp. And *H. uninervis*. Significant variations in the protein content of leaves and rhizome were observed in all the seagrass species (F=3.906 and 9.935, respectively, p<0.01). These results suggest that the rhizomes and shoots are the storage organs from which soluble carbohydrates and proteins can be mobilized for plant growth[25].

Lipid content in the seagrass was not a striking feature as it showed only very little difference between the species. Lipid content of seagrass leaves and rhizomes showed minimum in *T. hemprichii* and the maximum in *E. acoroides* and almost all the seagrass species recorded higher lipid content in rhizomes than the leaves. Significant variations in the lipid content of leaves and rhizomes were observed in all the seagrass species (F=10.098 and 16.384, respectively, p<0.01). Variations in the total lipid content of the seagrass species could be attributed to age, stage of growth and ecological variations[26]. Present study also found distinct seasonal variations in lipid content, registering the maximum in summer and the minimum in monsoon seasons.

Phenol content was higher both in leaves and rhizomes. It varied between 0.01 and 9.2 mg g^{-1} in all the species; the minimum was noted in *H. beccarii* and the maximum was recorded in the leaves of *E. acoroides*. Presence of much higher levels of phenol content in seagrass leaves has been reported earlier[27]. Significant variation in the phenol concentration of leaves and rhizomes was observed in the seagrass species (F=7.259 and 9.210, respectively, p<0.05). Such higher phenol content in seagrass leaves could be attributed to the defense mechanism of the plants against the epiphytes. It should be noted that leaves are much more exposed to epiphytes than the rhizomes, in general, rhizomes accumulate more phenol than the leaves, contrary present study recorded higher phenol content in leaves. Higher phenol content in rhizomes could be ascribed to the rhizome life span which is much longer than leaves, in additional to the environmental pressures[28].

Table 2—Calorific values of seagrasses, seaweeds and vegetables

Sl. No.	Plants		Energy (k Cal g^{-1})
1	*E. acoroides*#	Leaves	68.82
		Rhizomes	77.84
2	*H. beccarii*#	Leaves	29.16
		Rhizomes	38.84
3	*H. ovalis*#	Leaves	24.08
		Rhizomes	37.03
4	*C. rotundata*#	Leaves	60.62
		Rhizomes	63.68
5	*C. serrulata*#	Leaves	43.40
		Rhizomes	37.33
6	*H. uninervis*#	Leaves	40.15
		Rhizomes	37.16
7	*H. pinifolia*#	Leaves	28.39
		Rhizomes	28.23
8	*S. isoetifolium*#	Leaves	38.38
		Rhizomes	30.48
9	*Turbinaria conoides*$		23.98
10	*Sargassum wightii*$		22.46
11	*Gracilaria corticata*$		18.69
12	*Gracilaria edulis*$		19.41
13	*Gelidium pusillum*$		27.46
14	*Hypnea musciformis*$		12.08
15	Bengal gram leaves**		97.00
16	Cabbage**		27.00
17	Bitter gourd**		25.00
18	Urinal**		24.00
19	Cauliflower**		30.00
20	Drumsticks**		26.00
21	Jack Fruit, Tender**		51.00
22	Ladies Finger**		35.00
23	Peas**		93.00
24	Beetroot**		43.00
25	Carrot**		48.00
26	Onion**		59.00
27	Potato**		97.00
28	Sweet potato**		120.00

#seagrass, $seaweed (24) and **vegetable (29)

The two-way ANOVA has revealed the significant variation in tannin content of leaves and rhizomes (F=2.629 and 4.832, $p<0.05$), reflecting on the seasonal and inter-species variations in the tannin content. Earlier reports indicate that seagrasses are capable of accumulating significant levels of condensed tannins in leaf and shoot tissues[29]. However, difference in the concentration of tannins in different seagrass leaves remains unraveled and more importantly the tannin and phenol contents of seagrass leaves are much related to the defense mechanism of the seagrasses against epiphytic algal growth.

Calorific values of the seagrasses were found to be more in several species among them *E. acoroides* and *C. rotundata* are comparable to that of potato and sweet potato[29] (Table 2). Though the calorific values are appreciable in seagrass rhizomes and leaves, presence of phenols and tannins are the limiting factor in using these plants in regular diet. Such higher calorific contents of the seagrass leaves could be the reason for the giant marine mammals like sea cows and green turtles feed on them. Some of the important biochemical constituents laden in the seagrass tissues would be providing higher life span to these animals. So, further research is essential to isolate such compounds from the seagrasses for human use.

Conclusion

The calorific values are appreciable in seagrass rhizomes and leaves and presence of phenols and tannins is the only limiting factor in using these plants in human diet. Development of proper cooking methods for removing the phenols and tannins from seagrass tissues would help utilize theses plants as feed/food.

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