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Vitamin C content of some macroalgae of Visakhapatnam, east coast of India

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Vitamin C is analysed on monthly samples of 24 species of macroalgae collected for one year from the intertidal habitats of Visakhapatnam coast. Chlorophyll-*a* and carbohydrate are also analysed. In general, the algae belonging to Chlorophyceae and Phaeophyceae have higher annual mean contents of the vitamin (360 and 352 µg/g respectively) than Rhodophyceae (271 µg/g). The species which are rich in vitamin C are *Padina tetrastomatica* (513 µg/g), *Spongomorpha indica* (510) and *Chaetomorpha antennina* (488). The vitamin C maxima was observed during summer for Chlorophyceae and Phaeophyceae and during winter for Rhodophyceae. The vitamin C showed significant positive relationship with chlorophyll - *a* in Rhodophyceae and Chlorophyceae. It also showed a significant positive relationship with carbohydrate content of the latter. The distribution of vitamin C is associated with the morphogenetically important vegetative and reproductive phases indicating its key role in metabolic function.

There is a growing need to assess the food value of marine algae in view of their proven potential for use as food and fodder. The food value of algae comes not only from the bulk of nutrients provided, but also from the essential vitamins contained[1]. Vitamin C is an antioxidant and protects hydrogen/ electron carriers within the cell and maintains suitable redox levels for enzyme systems. Ascorbate is also involved in the biosynthesis of hormones and deoxyribose-nuclic acid (DNA). Several reports suggested that vitamin C occurs richly in most seaweeds[2-7]. However, since it has not been assayed so far in the algae of Visakhapatnam coast, we present its data in this paper, together with the data of chlorophyll - *a* and carbohydrate. The seasonal study made on vitamin C of different algae will fill the gap of knowledge for this area besides suggesting vitamin C rich species for possible exploitation.

Materials and Methods

Two stations are selected along the coastline of Visakhapatnam, st 1 at the red light house of VUDA park and st 2 at the Tenneti Park (Fig. 1) for sample collection. Both the stations are free from polluting effluents and have clear waters throughout the year except in the rainy season. Rocks of various sizes and rocky platforms, are exposed at the stations during low tide and offer a variety of habitats for the growth of algae. A total of 150 samples belonging to 24 species were collected from the intertidal zone at monthly intervals from May 1996 to April 1997. Of these species, 13 are annual and the rest are ephemeral. The 24 species of macroalgae of Visakhapatnam analysed belong to Chlorophyceae (12), Phaeophyceae (3) and Rhodophyceae (9). Most of the samples occur as lithophytes and a few as hydrophytes (submerged) and *Cladophora utriculosa* is an epiphyte. The plants are carefully isolated,

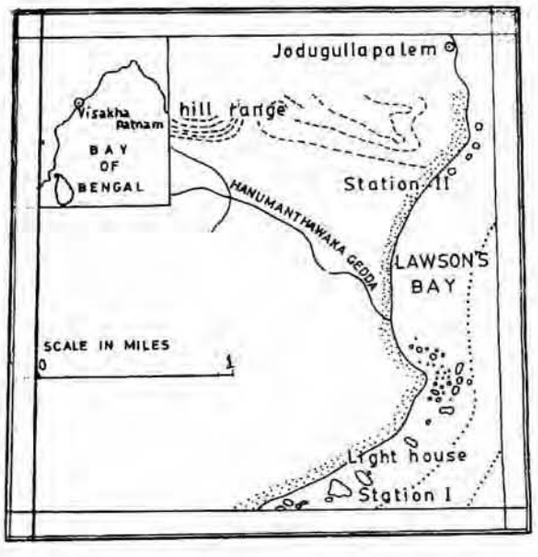

Fig. 1—Station location (83° 21′25″E 17°45′N) map of Visakhapatnam coast.

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cleaned to remove the extraneous matter, washed with seawater followed by fresh water in the laboratory and freezed. The frozen samples are dried in a freeze dryer (Martin Chryst) and powdered. These powders are used for analyses of vitamin C and carbohydrate. Chlorophyll – *a* was however, analysed on freshly collected wet samples of the first few months[8]. The carbohydrate estimation was done by the anthrone – sulfuric acid method[9] and vitamin C determination by the TCA (trichloroacetic acid) extraction procedure[10] which is briefly as follows.

To the powdered algal sample (*ca* 50 mg), 5% TCA (6 ml) was added, the contents shaken well and centrifuged to get a protein free extract of the vitamin. To a small portion (2 ml) of this clear solution, indophenol reagent (1 drop) which was prepared by dissolving 2,6-dichlorophenol indophenol (20 mg) in warm distilled water (10 ml) was added. The dinitrophenyl hydrazine and thiourea (DT) mix reagent (0.5 ml) prepared by dissolving 2,4-dinitrophenyl hydrazine (2 g) and thiourea (1 g) in 9 normal sulfuric acid (100 ml) was then added and the mixture was incubated at 60±1°C for 1 h. The reaction tube was then cooled in an ice water bath and 85% sulfuric acid (2.5 ml) was added to it while shaking. The red colour that developed was measured using a UV-2000 Spectrophotometer, Chemito at 540 nm after 30 minutes.

Results and Discussion

A summary of the biochemical results is shown in Table 1.

Chlorophyll-a — Chlorophyll - *a* is common in all the seaweeds. The total chlorophyll content was highest in Chlorophyceae (Table 1), being in the range of 11.8 µg/g in *Chaetomorpha brachygona* to 1.62 µg/g in *Ulva fasciata*. The pigment ranged from 7.56 µg/g in *Padina tetrastomatica* to 3.25 µg/g in *Sargassum tenerrimum* of Pheaophyceae and in Rhodophyceae from 1.92 µg/g in *Hypnea musciformis* to 0.47 µg/g in *Gracilaria corticata*. These values agree with those reported earlier[11].

Carbohydrates—The mean carbohydrate (Table 1) was higher in Rhodophyceae (43%) compared to Phaeophyceae (28) and Chlorophyceae (25). A carbohydrate maximum of 74.3% was recorded during December in *Pterocladia heteroplatos* and a minimum of 4.8% in February in *Amphiroa fragilissima*. Highest carbohydrate content was reported earlier[12,13] in Rhodophyceae and this may be attributed to the cellular enrichment of agarose related phycocolliods in these algae. The seasonal distribution of carbohydrate showed a peak in winter (October-February) in most of the seaweeds except in *Gracilaria* (February-April) and *Amphiroa* (August). Similar peak for carbohydrate during winter was also reported for algae of the Goa coast[14].

Vitamin C— The content of vit - C as annual mean (Table 1) was higher in Chlorophyceae (360 µg/g) than in Phaeophyceae (352 µg/g) and Rhodophyceae (210 µg/g). Unlike Chlorophyceae and Phaeophyceae, Rhodophyceae do not synthesize water-soluble vitamins and this might be the reason for the low content observed in this group[15]. Among the seaweeds analysed the submerged algae such as *Padina tetrastomatica* and *Spongomorpha indica* yielded higher vit - C than the littoral species. Even though higher content is observed in *Cladophora utriculosa* it occurs as a seasonal epiphyte and could be sampled only once, hence the former are considered as better yielding species. Species such as *Pterocladia heteroplatos*, *Ulva fasciata*, *Liagora erecta*, *Gelidium pusillum* and *Gracilaria corticata* yielded the least (Fig. 2). In algae of the Adriatic[6] and Mediterranean[7] coasts, brown algae are reported to have higher vitamin content. From eleven species of the Gulf of Mannar[2], and from *Ulva fasciata* of Gujarat[4] coast the vitamin was reported at less concentrations. However, as the analysis done was of wet samples, no comparison can be made with the present study. The variations in vit - C content may be attributed not only to the test organism but also to the ecological and geographical factors[6].

The seasonal variation in vit - C of 13 algae that

Table 1—Chemical composition of macroalgae.

Class	Range	Mean
	Vitamin – C (µg/g)	
Chlorophyceae	89.6 – 995.2	360.38 ± 9.7
Phaeophyceae	140.8 – 932.8	352.30 ± 68.19
Rhodophyceae	3.2 – 587.2	210.51 ± 16.4
	Carbohydrate (%)	
Chlorophyceae	16.1 – 41.7	25.1 ± 0.012
Phaeophyceae	10.0 – 57.6	27.9 ± 0.559
Rhodophyceae	4.8 – 74.3	43.15 ± 0.477
	Chlorophyll (µg/g)	
Chlorophyceae	0.288 – 14.1	7.96 ± 0.170
Phaeophyceae	3.48 – 9.47	6.296 ± 0.325
Rhodophyceae	0.225 – 3.61	1.386 ± 0.064

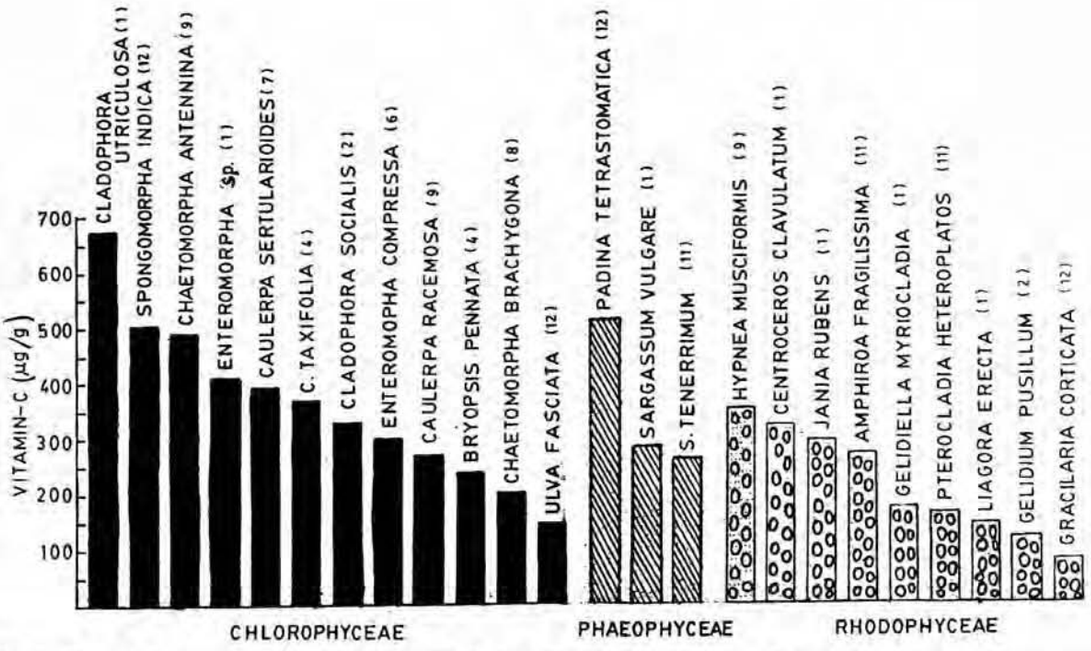

Fig. 2—Vitamin C concentration in different groups of algae. Figures in parenthesis indicate the number of samples analysed.

are occurring continuously is shown in Fig. 3. The content of vit - C in most seaweeds varied markedly with the seasons. Most of the species showed two peaks with one corresponding to the vegetative part of early growth phase and the other to the reproductive phase of the thalli. Such phases observed in *Ulva fasciata* are one in August and other in March. *Enteromorpha compressa* which is analysed only for six months displayed a peak in February, *Chaetomorpha antennina* in September and November, *C. brachygona* in August and February, *Caulerpa racemosa* in September and February, *C. sertularioides* in August and December, and *Spongomorpha indica* in September and December. Among Phaeophyceae *Padina tetrastomatica* showed two peaks, one in May and the other in January and *Sargasssum tenerrimum* in June and December. Among Rhodophyceae, *Gracilaria corticata* showed two peaks, one in September and the other in February to March. No such distinctions are observed in *Hypnea musciformis*. In *Pterocladia heteroplatos*, the first peak was in June and the other in January and in *Amphiroa fragilissima* one in June and the other in October. From the group-wise seasonal mean variation (Table 2), it is evident that taxa of Chlorophyceae and Phaeophyceae showed summer maxima and Rhodophyceae winter maxima. Seasonal

Table 2—Seasonal mean variation in vitamin C (µg/g) of macroalgae of Visakhapatnam

Class	Summer*	Monsoon**	Winter***
Chlorophyceae	344.1 ± 7.19	335.2 ± 6.5	329.3 ± 6.5
Phaeophyceae	450.6 ± 29.7	330.2 ± 14.2	371.9 ± 19.0
Rhodophyceae	165.0 ± 5.4	187.7 ± 9.1	243.8 ± 9.5

*February to May
**June to September
***October to January

variations in ascorbic acid content are apparently dependent on hydrographic parameters and solar irradiation on one hand and the growth intensity of plants on the other[6]. The activity of ascorbic acid in regulating cell metabolism and growth is however due to its ability to undergo oxidation – reduction processes.

Algae with brighter thalli such as *Cladophora utriculosa, Spongomorpha indica, Cheatomorpha, antennina, Padina tetrastomatica* and *Hypnea musciformis* were found to be rich in vit - C and this may be because of the vitamin stimulates the healthy growth of the plant[7] being an electron acceptor and performs the role of an antioxidant and protects the algae[10]. In younger parts of thalli, ascorbic acid dominates over its oxidized form[5]. The vitamin seems

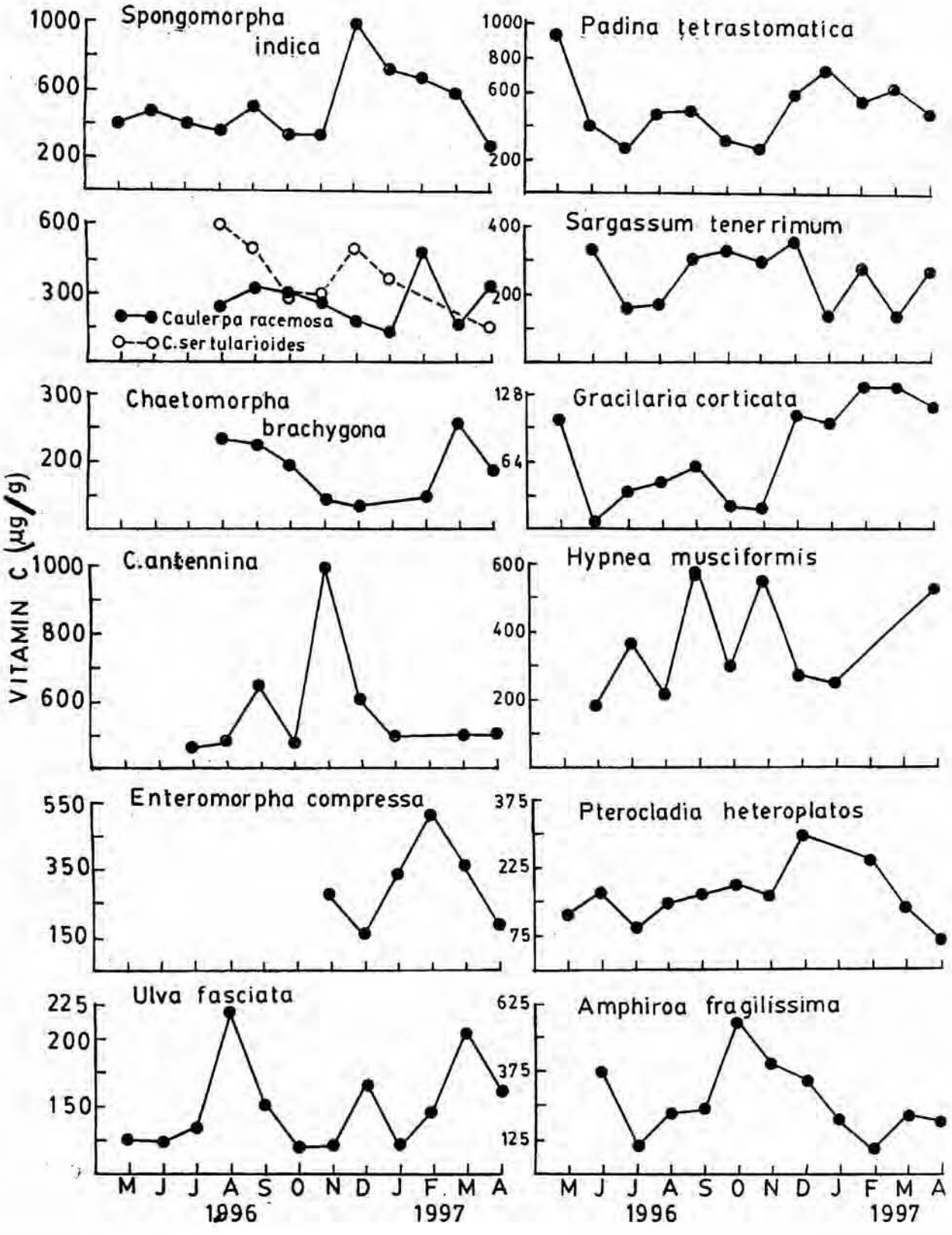

Fig. 3—Seasonal variation in vitamin C content of continuously occurring algal species.

to have a significant (p>99%) linear relationship with chlorophyll *a* in red algae (Eq. 1) and green algae (Eq. 2).

Vit C =56.2 + 99.5 (Chl - *a*), r = 0.78 (n=16) ...(1)

Vit C =119.7 + 30.9 (Chl-*a*), r = 0.83 (n=17) ...(2)

This may be because the vitamin is enriched in organogenetic centres of plant species during differentiation[16]. The distribution of vit - C on chl - *a* of brown algae is however scattered and did not indicate any association. The vit – C seemed positively associated with carbohydrate of green algae in which it was low by the regression equation which is significant at p>99% due to high value of 'n'

Vit C = –51.3 + 15.5 (Carbohydrate),
r = 0.44 (n=51) ...(3)

No significant relationship existed between vit - C and carbohydrate for the members of Rhodophyceae and Phaeophyceae. They have reserve polysaccharides based on galactose and mannose respectively, which cannot, however, meet the requirement for vit - C production as the biosynthesis of vit - C takes place from D-glucose[17]. *Padina tetrastomatica* seems to be an exception since vit-C content increased steadily with carbohydrate, except when the vitamin itself was at the peak (933 µg/g) in May and when the carbohydrate reached its peak (37.4%) in November.

In conclusion the vit - C content of the marine algae varies markedly with season, differently for each group and species. The submerged species are richer in vit - C irrespective of their taxonomical grouping. Maxima of the vitamin are observed during summer for Chlorophyceae and Phaeophyceae and in winter for Rhodophyceae. The vit - C seemed to influence the health of the plants as brighter algae are observed to have higher content of it.

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References

1 Mumford Jr.T F & Miura A, in *Algae and human affairs*, edited by C A Lembi & J R Waaland, (Cambridge University Press, Cambridge) 1988, 91.
2 Thivy F, in *Proc Symp Algology*, (ICAR, New Delhi) 1960, 345.
3 Jensen A, *Proc Int Seaweed Symp*, 4 (1964) 319.
4 Subbaramaiah K, *Phykos*, 6 (1967) 115.
5 Liso R, Calabrese G & Chieppa M, *Phycologia*, 17 (1978) 1453.
6 Munda I M, *Hydrobiologia*, 151/152 (1987) 477.
7 Shaalan S H, Khaleafa A F & Kareem M S A, *Bull Nat Inst Ocenogr & Fish* (Egypt), 17 (1991) 57.
8 Jeffrey S W & Humphrey G F, *Biochem Physiol Pflanz*, 167 (1975) 191.
9 Parson T R, Maita Y & Lalli C M, *A manual of chemical and biochemical methods for sea water analysis*, (Pergamon Press, London) 1984, 173.
10 Baker H & Frank O, *Clinical vitaminology: Methods and interpretations*, (Wiley, New York) 1968, pp. 198.
11 Reeta Jayasankar & Ramalingam J R, *Seaweed Res Util*, 16 (1993) 41.
12 Chennubhotla V S K, Najmuddin M, Ramalingam J R & Kaliaperumal N, *CMFRI Bulletin*, 44 (1990) 16.
13 Dhargalkar V K, Jagatap T G & Untawale A G, *Indian J Mar Sci*, 9 (1980) 297.
14 Sumitra Vijayaraghavan, Rajagopal M D & Wafar M V M, *Indian J Mar Sci*, 9 (1980) 61.
15 Jayasree V, Solimabi & Kamat S Y, *Indian J Mar Sci*, 14 (1985) 228.
16 Chinoy J J, *Indian J Plant Physiol*, 10 (1967) 202.
17 Gruen M & Loewus F A, *Planta*, 160 (1984) 6.