The biochemical composition of *Enteromorpha* spp. from the Gulf of Gdańsk coast on the southern Baltic Sea*

OCEANOLOGIA, 42 (1), 2000.
pp. 19–28.

KEYWORDS
Enteromorpha spp.
Macroalgae
Biochemical composition

Amany Mohamed Haroon
National Institute of Oceanography
and Fisheries,
Academy of Scientific Research
and Technology,
101 Kaser El-eni St., Cairo, Egypt

Anna Szaniawska,
Monika Normant,
Urszula Janas
Institute of Oceanography,
University of Gdańsk,
al. Marszałka Piłsudskiego 46, 81–378 Gdynia, Poland; e-mail: oceasz@univ.gda.pl

Manuscript received 16 August 1999, reviewed 21 January 2000, accepted 31 January 2000.

Abstract

Variations in lipid, protein and carbohydrate contents of *Enteromorpha* spp. were examined over a seven-month period from April to October 1993. The samples were collected from seven sampling stations along the Gulf of Gdańsk coast. The lipid content was low and varied slightly from $3.47 \pm 1.76\%$ of DW at Puck to $4.36 \pm 2.17\%$ of DW at Rewa and Chałupy. The protein content varied from $9.42 \pm 4.62\%$ of DW at Puck to $20.60 \pm 5.00\%$ of DW at Jurata. At the remaining stations the values vary over a narrow range. The maximum protein contents were recorded at the beginning and end of the growing season. The level of carbohydrate was very high compared to that of lipid and protein and varied from $29.09 \pm 6.44\%$ of DW at Osłonino to $39.81 \pm 11.15\%$ of DW at Puck. Seasonal carbohydrate changes were noted at all sampling stations, the minimum occurring in spring and autumn and the maximum in summer.

* This research was supported by grant BW/1320–5–0169–9 from the University of Gdańsk.

1. Introduction

Enteromorpha spp. are currently the dominant group of algae in the littoral zone of the Gulf of Gdańsk (Pliński *et al.* 1988). The importance of this genus in the biocenosis of the Gulf of Gdańsk has risen as a result of the declining numbers of other taxa, such as *Fucus vesiculosus* and *Furcellaria fastigata*, possibly because of changing environmental conditions and the ongoing eutrophication of the study area. Regarding the economic importance of *Enteromorpha* spp., the presence of a great variety of essential amino acids has been recorded in *Enteromorpha intestinalis*. It also has a good capacity for fermentation, which is shown by methanisation tests (Sauze 1981). Moreira-da-Silva *et al.* (1982) studied the possibility of using marine macrophytes as a substrate for biogas generation. Special research attention has been given to *Enteromorpha* spp. and *Ulva* spp. as a source of methane generation through anaerobic digestion, the time involved in gas production and the possibility of using the depleted biomass as a fertiliser for phytoplankton cultures. Finally, the possibility of using algae for other energetic options has been analysed and their potential compared with that of the water hyacinth *Eichornia crassipes*.

Enteromorpha spp. have also been used as a source of bioactive compounds similar to those which cause an inhibitory effect against the bacterium *Xanthomonas oryzae*, which causes leaf blight disease in paddy crops (Manimala & Rengasamy 1993). People in the Philippines and Japan also use *Enteromorpha* spp. as food (Hoppe 1966, Tamura 1970, Velasquez 1972). There is a lack of information concerning the biochemical composition of *Enteromorpha* spp. from the Gulf of Gdańsk. Studies have been done on the relation between heavy metal accumulation and the biochemical composition of *Enteromorpha* spp. from the Gulf of Gdańsk, and one study addressed its energy value and lipid content (Haroon & Szaniawska 1994, Haroon & Szaniawska 1995).

The aim of the present study was to determine the nutritional value of *Enteromorpha* spp. by identifying their biochemical composition (lipid, protein and carbohydrate). It was also of interest to determine when and where these components were at their maximum and minimum. Therefore, close attention was paid to geographical and seasonal changes.

2. Material and methods

Samples of *Enteromorpha* spp. were collected at monthly intervals from April to October 1993 at seven sampling stations distributed along the western part of the Gulf of Gdańsk coast (Fig. 1). The algal samples were

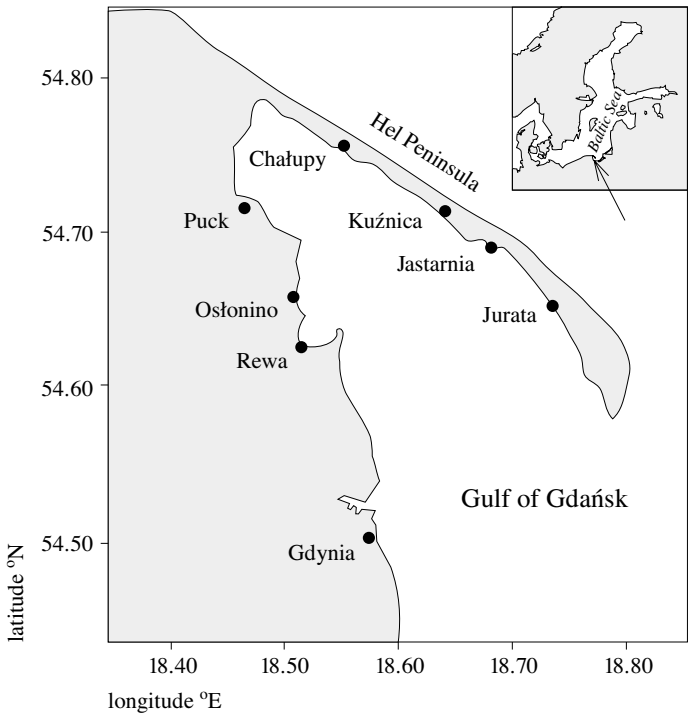

Fig. 1. Map of the Gulf of Gdańsk with sample collection points marked

hand picked from shallow littoral water. At each station three sub-samples were taken and analysed separately. In the laboratory, they were cleaned, washed with distilled water and dried at 55°C to constant weight. Then they were powdered, sifted through a piece of muslin and stored for analysis.

A 1:2 mixture of chloroform : methanol was used for lipid extraction (Bligh & Dyer 1959). The total lipid content was measured as described by Marsch & Weinstein (1966). The results obtained were recalculated on the basis of a standard curve prepared for glyceryl tripalmitate.

The total protein content was determined with Folin reagent, with bovine albumin serving as the standard (Lowry *et al.* 1951).

The total carbohydrate content was assayed by the phenol–sulphuric acid method (Dubois *et al.* 1956), which involved extraction with 15% trichloroacetic acid. The results were calculated from a glucose standard curve. Lipid, protein and carbohydrate contents were expressed as the percentage dry weight (% of DW). The results are given as a mean with standard deviation (± SD). A comparison of mean values (*t*-test) was used for the statistical calculations.

Table 1. The minimum, mean (± SD) and maximum contents of lipid, protein and carbohydrate in *Enteromorpha* spp. from the Gulf of Gdańsk during the period from April to October 1993

Station	Number of samples	Lipid (% of DW)			Protein (% of DW)			Carbohydrate (% of DW)		
	n	min	mean ± SD	max	min	mean ± SD	max	min	mean ± SD	max
Rewa	5	2.62	4.36 ± 2.17	7.77	6.27	12.14 ± 5.26	20.13	13.61	32.46 ± 5.26	65.11
Osłonino	6	2.71	4.06 ± 1.07	5.25	4.16	13.78 ± 7.24	20.81	18.63	29.09 ± 6.44	34.86
Puck	5	1.88	3.47 ± 1.76	5.39	5.60	9.42 ± 4.62	17.20	27.67	39.81 ± 11.15	54.71
Chałupy	6	2.43	4.36 ± 1.85	7.13	12.22	16.52 ± 3.95	21.61	20.05	32.52 ± 11.01	45.32
Kuźnica	6	2.31	3.86 ± 1.78	6.39	5.54	12.94 ± 7.43	21.33	19.60	32.03 ± 11.73	46.94
Jastarnia	6	2.72	3.64 ± 1.04	5.53	6.64	15.25 ± 7.99	27.37	20.92	37.45 ± 9.09	48.78
Jurata	7	2.40	3.72 ± 1.34	6.02	15.89	20.60 ± 5.00	28.74	25.15	34.81 ± 12.70	53.75

DW – dry weight, SD – standard deviation, min – minimum value, max – maximum value.

3. Results

The average lipid, protein and carbohydrate values of the *Enteromorpha* spp. samples collected at seven sampling stations distributed along the western coast of the Gulf of Gdańsk are presented in Table 1.

The lipid content in *Enteromorpha* spp. from the Gulf of Gdańsk was generally low and showed very few geographical changes (Fig. 2). In most months, these changes were only about 1% of DW, except during June 1993 when the changes in values ranged from $2.31 \pm 0.29\%$ of DW at Kuźnica to $7.77 \pm 0.97\%$ of DW at Rewa. At all stations the maximum lipid content was noted in spring, at the beginning of the growing season (Fig. 3a). After this period, the values decreased and remained nearly stable.

The protein content of *Enteromorpha* spp. varies geographically. The seasonal changes, with the maximum in spring and autumn (at the beginning and end of the growing season) and the minimum in summer, are shown in Fig. 3b. During the investigated period, the protein content of *Enteromorpha* spp. from the Gulf of Gdańsk ranged from 9.42 ± 4.62 to $20.60 \pm 5.00\%$ of DW.

As was the case with protein content, the carbohydrate content of *Enteromorpha* spp. showed geographical variation. The mean values for all samples ranged from $29.09 \pm 6.44\%$ of DW at Osłonino to $39.81 \pm 11.15\%$ of DW at Puck (Table 1). The minimum carbohydrate content was noted in April and October 1993, the maximum in August 1993 (Fig. 3c). The exception was Puck, where the maximum value ($54.71 \pm 8.17\%$ of DW) was in July.

4. Discussion

In comparison to protein and carbohydrate, lipid makes up the relatively smallest proportion in *Enteromorpha* spp. No statistically significant changes in lipid content between sampling stations could be detected ($p > 0.05$). Seasonal variations in lipid content, with the maximum in spring, were observed at all sampling stations. Most of this lipid was tightly bound to the wall fabric. Temperature has a characteristic effect on many plant lipids in that it increases the level of unsaturation of acyl chains, which slows down both metabolism and transport (Jones & Harwood 1993). Therefore, during the period of this study the maximum lipid content of *Enteromorpha* spp. was found at the beginning of the growing season in spring. Consequently, as the temperature increased, the lipid level decreased and remained almost stable until the end of the growing season. Dodson & Aronson (1978) noted that lipid accounted for nearly 14% of the cell wall weight of *E. intestinalis*. The results obtained are slightly higher than

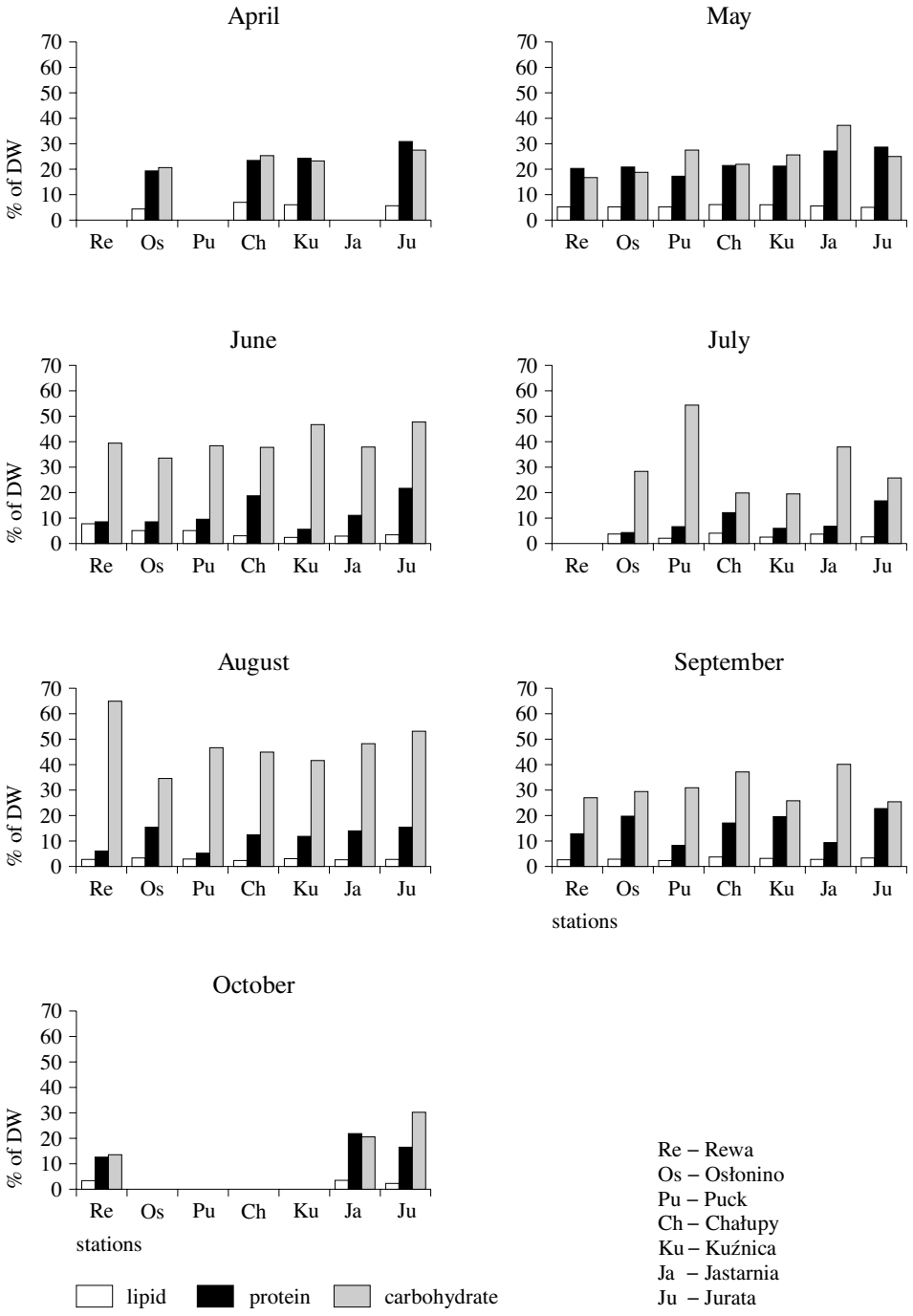

Fig. 2. Variations in lipid, protein and carbohydrate contents (% of DW) in *Enteromorpha* spp. from different stations in the period April–October 1993

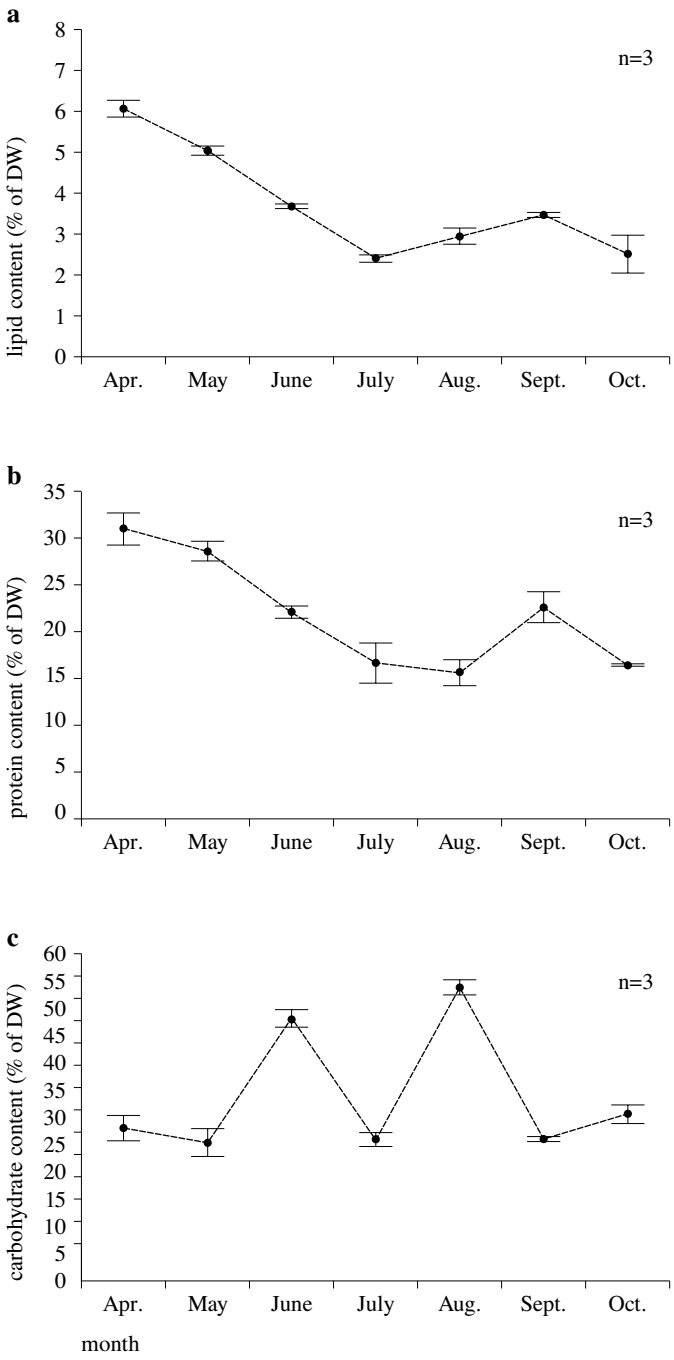

Fig. 3. Seasonal changes in the lipid (a), protein (b) and carbohydrate (c) contents (% of DW) of *Enteromorpha* spp. from Jurata. Values are given as a mean with standard deviation

those of Banaimoon (1992) for *Enteromorpha flexuosa* from southern Yemen (4.6%), and similar to those of the same author for *Codium temontosum* and *Halimeda tuna*. However, they are higher than the values of *Ulva* spp. and species related to *Phaeophyta* and *Rhodophyta*.

Protein is a significant building component, so variations in its content are not very high. In *Enteromorpha* spp. from the Gulf of Gdańsk the content depended on location. During the whole study period, the minimum protein content was found at Puck ($9.42 \pm 4.62\%$ DW) and the maximum at Jurata ($20.60 \pm 5.00\%$ of DW). At the same time, similarity between some stations was noted. Although there is not enough information concerning the nutrient level at each station, it is suspected that the changes can be related to differences in this feature, as well as to changes in species composition of the material examined. The cycles of seasonal changes in protein content were observed at all sampling stations and, presumably, were related to requirements for structural tissues. This is inferred from the fact that the maximum protein content was recorded at the beginning of the growing period in spring (the period of slowest growth) and again at the end of the growing season in autumn. The minimum protein content was noted in summer. The values recorded are similar to those reported by Munda & Gubensek (1976, 1986), Owens & Stewart (1983), Tkachenko & Koval (1990) and Wheeler & Björnsäter (1992).

Changes in carbohydrate content at the different sampling stations were observed throughout the study period. Carbohydrate is the most important component for metabolism as it supplies the energy needed for respiration and other metabolic processes. During summer, after the period of maximum growth had occurred, the carbohydrate content was higher than it had been in spring and showed a tendency to fluctuate. The fact that the water temperature in the Gulf of Gdańsk is highest during the summer months, together with the high solar radiation during that time, makes for optimal photosynthesis conditions. This process may well lead to an increase in the carbohydrate concentration. It could be that the annual changes in carbohydrate content are dependent on a diverse species composition, as well as on the different environmental conditions obtaining at each station.

5. Conclusions

- The quantitative composition of *Enteromorpha* spp. shows that the major constituent is carbohydrate, followed by protein and lipid.
- The lipid content is low and remains almost stable throughout the year, ranging from $3.47 \pm 1.76\%$ of DW at Puck to $4.36 \pm 2.17\%$ of DW at Rewa and Chałupy.

- The protein content varies seasonally and geographically. The average values ranged from $9.42 \pm 4.62\%$ of DW at Puck to $20.60 \pm 5.00\%$ of DW at Jurata, with the maximum at the beginning and end of the growing season.
- Carbohydrate contributes the relatively largest proportion of the biochemical composition. Its values vary considerably with sampling station and season.
- The nutritional value of *Enteromorpha* spp. is higher during spring and autumn (high protein and lipid content) than during summer, when their carbohydrate content is high.

Acknowledgements

We would like to thank an anonymous reviewer for improving an earlier version of this manuscript. The linguistic assistance of Jennifer Zielińska is gratefully acknowledged.

References

Banaimoon S. A., 1992, *Fatty acids in marine macroalgae from Southern Yemen (Hadramout) including occurrence of eicosatetraenoic (20:4) and eicosapentaenoic (20:5) acids*, Bot. Mar., 35, 165–168.

Bligh E. G., Dyer W. J., 1959, *A rapid method of total lipid extraction and purification*, Can. J. Biochem. Physiol., 37, 911–917.

Dodson J. R., Aronson J. M., 1978, *Cell wall composition of Enteromorpha intestinalis*, Bot. Mar., 21, 241–246.

Dubois M., Gilles K. A., Hamilton J. K., Rebers P. A., Smith F., 1956, *Calorimetric methods for determination of sugars and related substances*, Analit. Chem., 28, 350–356.

Haroon A. M., Szaniawska A., 1994, *Relation between biochemical composition of Enteromorpha spp. and heavy metal accumulation. Abstract of Biochemical and Physiological effects of Pollutants and Toxicological Assessment of Environmental of quality*, Annual Conference in Genova (Italy), Sept. 20–23.

Haroon A. M., Szaniawska A., 1995, *Variations in energy values and lipid content in Enteromorpha spp. from the Gulf of Gdańsk*, Oceanologia, 37 (2), 171–180.

Hoppe H. A., 1966, *Nahrungsmittel aus Meeresalgen*, Bot. Mar., 9 (Suppl.), 18–40.

Jones A. L., Harwood J. L., 1993, *Lipids and lipid metabolism in the marine alga Enteromorpha intestinalis*, Phytochemistry, 34 (4), 969–972.

Lowry O. H., Farr A. L., Randall R. J., Rosebrough N. J., 1951, *Protein measurement with Folin phenol reagent*, J. Biol. Chem., 193, 265–275.

Manimala K., Rengasamy R., 1993, *Effect of bioactive compounds of seaweeds on the Phytopathogen Xanthomonas oryzae*, Phykos., 32 (1)–(2), 77–83.

Marsch J. B., Weinstein D. B., 1966, *Simple charring method for determination of lipids*, J. Lipid Res., 7, 574–576.

Moreira-da-Silva P. C., Bastos-Netto D., Costa-Muniz A., Nobre-Barros Y., 1982, *Algae: Energetic utilization in Brazil*, Pub. Inst. Pesqi. Mar., 146, 8–17.

Munda I. M., Gubensek F., 1986, *The amino acid composition of some benthic marine algae from the Northern Adriatic*, Bot. Mar., 29, 367–372.

Munda I. M., Gubensek F., 1976, *The amino acid composition of some common marine algae from Iceland*, Bot. Mar., 19, 85–92.

Owens N. J. P., Stewart W. D., 1983, *Enteromorpha and the cycling of nitrogen in a small estuary*, Estuar. Coast. Shelf Sci., 17 (3), 287–296.

Pliński M., Florczyk I., Galińska M., 1988, *The taxonomy of the genus Enteromorpha Link in the Gulf of Gdańsk. A numerical approach*, Kieler Meeresforsch. Sonderh., 6, 265–271.

Sauze F., 1981, *Chemical and energetic potential of aquatic biomass*, Tech. Eau. Assain., 413, 7–23.

Tamura T., 1970, *Marine aquaculture*, Nat. Sci. Foundation PB 194 OSIT, Part II, Washington D. C.

Tkachenko F. P., Koval V. T., 1990, *Biochemical composition of abundant benthic seaweeds of the Black Sea*, Hydrobiol. J., 26 (6), 39–43.

Velasquez G. T., 1972, *Studies and utilization of the Philippine marine algae*, Proceedings 7th International Seaweed Symposium, Univ. Tokyo Press, Tokyo, 62–65.

Wheeler P. A., Björnsäter B. R., 1992, *Seasonal fluctuations in tissue nitrogen, phosphorus and N:P for five macroalgal species common in the Pacific northwest coast*, J. Phycol., 28, 1–6.