Effect of Different Environmental Factors on the Major Chemical Constituents of Some Marine Algae

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ABSTRACT. An investigation of the effect of 15 environmental factors on ash, crude fibre, lipids, protein and carbohydrate content of *Enteromorpha intestinalis* (green), *Sargassum salicifolium* (brown) and *Gelidium latifolium* (red) revealed that these chemical constituents were influenced by several environmental factors. On the other hand, the effect of these environmental factors was species – specific.

In general, total soluble salts, nitrite, nitrate, potassium, inorganic phosphorus, pH and water temperature were the major controlling factors; other factors were merely contributory.

Introduction

Several factors undoubtedly play a part in the causation of the fluctuation in the chemical composition of marine algae. According to Macpherson and Young (1952) the fundamental one is the metabolic activity but this is probably controlled by the temperature of the surrounding water, the concentration of essential nutrients and the intensity of light.

In the search of the reasons for fluctuations in composition of marine algae, numerous authors (El-Naggar 1980, Whyte and Englar 1980, Gorham and Lewey 1984, Rosell and Srivastava 1985, Zavednik 1987 and Sway *et al.*, 1991), studied the effect of seasons of the year on the chemical composition. However, few attempts (Fadeel *et al.*, 1981, Ramus 1983, Duke *et al.*, 1986 and Pettitt *et al.*, 1989) have been made to correlate other environmental factors with the chemical constituents of marine algae. At the same time, these studies have been confined to a limited number of the environmental factors.

In the work presented here, the author has studied, in the field, the effects of 15 environmental factors on ash, crude fibre, lipids, protein and carbohydrate content of *Enteromorpha intestinalis* (green), *Sargassum salicifolium* (brown) and *Gelidium latifolium* (red) which grow commonly on the Mediterranean coast of Egypt. This may provide the necessary information as regards the appropriate environmental conditions of the algal species for the production of one or more of their constituents.

Materials and Methods

Over a full year, regular seasonal visits were made to Abu-Qir - 24 km east of Alexandria - on the Mediterranean coast of Egypt. From this locality, the algal materials and water samples were taken. Sampling was continued from spring 1991 to winter 1992.

Algal Materials

The algae used in this work were: *Enteromorpha intestinalis* (L.) Link, *Sargassum salicifolium* (Bert.) J. Ag. and *Gelidium latifolium* (Grev.) et Thur. The algal nomenclature used is that recommended by El-Naggar (1980).

The algal samples were cleaned from epiphytes and non-living matrix in running water and rinsed many times in distilled water. The samples were then spread on string nets and allowed to dry in air. Air-dried samples were ground and stored in stoppered bottles at room temperature.

All samples were analysed for their ash, crude fibre, lipid, protein and carbohydrate content. Ash content was determined by following the method of Rosell and Srivastava (1985). Crude fibre content was determined according to the method described by El-Naggar (1980). Lipid content was extracted by chloroform-methanol mixture (2:1 v/v) following the method of Holme and Hazel (1983). Protein and carbohydrate were determined spectrophotometrically according to Coombs *et al.* (1987) and Dubois *et al.* (1956) respectively. All analyses were done in triplicate and the mean values were recorded.

Water Samples

Water samples were regularly obtained by one litre wide mouth graduated bottles. Each sample was a composite of 4 random samples from the collection site. At the time of sampling, the water temperature was recorded. The pH of water samples was determined by a portable pH-meter. The containers were kept in dark, packed on ice and carried to laboratory where the chemical analyses were carried out.

Chemical analyses were carried out on GF/C filtered water. Unless otherwise stated, the chemical parameters listed in Table 1 were analysed according to the methods described in APHA (1965 and 1985). All analyses were done in triplicate, and the mean values were recorded.

Results

Environmental Factors

The range and average of water temperature, pH, total soluble salts, total alkalinity, organic carbon, sulphate, silicate, inorganic phosphorus, nitrate, nitrite, ammonia and elements (Na, K, Ca and Mg) are tabulated in Table 1. Seasonal variations are shown in Fig. 1-3.

Water Temperature

Fluctuation in values of water temperature showed clear seasonality (Fig. 1a). The water temperature ranged between 14.0°C and 29.5°C. The highest value was measured in summer and the lowest in winter.

pН

During the period of investigation, the pH values of seawater were more or less within the alkaline range (Table 1). From Fig. (1b), it is apparent that highest value of pH (8.73) was recorded in autumn and the lowest (7.60) in summer.

Total Soluble Salts (analysed according to Jackson 1962)

Total soluble salts ranged between 35.52 and 4.2.00 g/l, with remarkable seasonal variations. The highest concentration was recorded in summer and the lowest in winter (Fig. 1c).

Total Alkalinity

Values fluctuated within a wide range (0.27-0.60 mg/l) with an average of 0.38 mg/l. 1. The highest concentration was recorded in spring while the lowest was obtained in autumn (Fig. 1d).

Organic Carbon (Piper, 1947)

Organic carbon content also showed a clear seasonal trend with high value (1.11 g/) 1) in autumn and low value (0.06 g/l) in spring (Fig. 2a)

Sulphate

From Table 1, it is apparent that amount of sulphate in seawater at Abu Qir locality did not differ greatly throughout the period of investigation. The difference between the maximum and the minimum was 1.87 g/l. The maximum value was recorded in summer while the minimum was obtained in spring (Fig. 2 b).

Silicate

Variations in silicate were large (Table 1 and Fig. 2c). The concentrations of silicate ranged from 8.23 μ g/l in spring to 28.80 μ g/l in autumn.



FIG. 1. Seasonal variations in temperature, pH, total soluble salts and total alkalinity content of surface seawater at Abu-Oir.

| TABLE 1. | Range and average of temperature, pH, total soluble salts, total alkalinity, organic carbon, sulphate, silicate, inorganic phos- | | | | | |
|----------|--|--|----------|--|--|--|
| | phorus, inorganic nitrogen and elements of surface seawater at Abu-Qir. | | | | | |
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| Environmental factor | Range | Average |
|------------------------------|----------------------------|--------------|
| Water temperature (°C) pH | 14.0 - 29.5 7.60 - 8.73 | 21.3 8.17 |
| Total soluble salts (g/l) | 35.52 - 42.00 | 38.66 |
| Total alkalinity (mg/l) | 0.27 - 0.60 | 0.38 |
| Sulphate (g/l) | 2.25 - 4.12 | 3.17 |
| Silicate (µg/l) | 8.23 - 28.80 | 18.09 |
| Inorganic phosphorus (µg/l) | 3.84 - 51.40 | 21.10 |
| Inorganic nitrogen : | | |
| Nitrate - N (mg/l) | 0.126 - 0.207 | 0.154 |
| Nitrite - N (mg/l) | 0.076 - 0.390 | 0.194 |
| Ammonia - N (mg/l) | 0.037 - 1.262 | 0.364 |
| Elements: | | |
| Sodium (g/l) | 9.70 - 16.20 | 12.95 |
| Potassium (g/l) | 0.39 = -0.85 | 0.67 |
| Calcium (meq./l) | 14.00 - 46.94 | 23.86 |
| Magnesium (meq./l) | 14.77 - 101.01 | 72.15 |

Inorganic Phosphorus

As indicated in Table 1, the inorganic phosphorus of seawater fluctuated within a wide range with an average of $21.10 \,\mu$ g/l. Values showed a noticeable seasonal trend. The lowest value was recorded in summer and the highest in winter (Fig. 2d).

Inorganic Nitrogen (nitrate -N, nitrite -N and ammonia -N)

From the results obtained for inorganic nitrogen, we find that throughout the period of investigation, the pattern of changes in inorganic nitrogen was greatly different (Fig. 3a&b).

As indicated in Fig. 3a, a lower range of fluctuations was recorded in nitrate-N (analysed according to Golterman 1969) levels. The range of values was from 0.126 mg/l in autumn to 0.207 μ /l in winter.

In contrast, ammonia-N (Solorzono 1969) and nitrite-N (Barnes and Folkard 1951) in seawater showed remarkable seasonal variations (Fig. 3a&b). The range in values for ammonia-N was 0.037 to 1.262 mg/l. The maximum occurred in autumn and the minimum in spring. On the other hand, the maximum value of nitrite-N was 0.390 mg/l in winter while the minimum was 0.76 mg/l in summer.

Further examination of the results (Table 1 & Fig. 3a&b) clearly shows that surface seawater at Abu-Qir was found to have considerably higher values of nitrite-N



FIG. 2. Seasonal variations in organic carbon, sulphate, silicate, and inorganic phosphorus content of surface seawater at Abu-Qir.

and ammonia-N especially throughout autumn and winter. This abnormal trend might be due to disposal of effluents into the sea. This locality receives considerable amounts of untreated effluents rich in organic matter.

Elements (Na, K, Ca & Mg)

Concentrations of sodium, potassium, calcium and magnesium in seawater were characterized by large seasonal variations (Fig. 3c-e).

Concentrations of Na and Mg showed similar fluctuations during the period of study. The highest concentrations were analysed in autumn and the lowest in spring (Fig. 3d&e).

Further consideration of Fig. 3(d&e) revealed that pattern of changes in K was, in general, similar to that of Ca. The highest values were recorded in spring while the lowest were obtained in winter.

Chemical Constituents

The range and average of ash, total lipids, crude fibre, protein and carbohydrate levels of *Enteromorpha intestinalis*, *Gelidium latifolium* and *Sargassum salicifolium* are shown in Table 2.

| Chemical constituent | E. intestinalis | | S. salicifolium | | G. latifolium | |
|----------------------|-----------------|---------|-----------------|---------|---------------|---------|
| | Range | Average | Range | Average | Range | Average |
| Ach | 20.20 - 56.17 | 36 77 | 18 20 - 30 41 | 23.81 | 12 50 - 18 60 | 14.47 |
| Crude fiber | 2.90 - 7.73 | 5.13 | 3.11 - 9.40 | 6.86 | 8.80 - 12.17 | 10.36 |
| Lipids | 1.17 - 4.82 | 2.64 | 0.36 - 2.90 | 1.63 | 1.00 - 2.85 | 1.73 |
| Protein | 2.28 - 10.16 | 2.88 | 3.66 - 8.65 | 5.69 | 1.59 - 6.63 | 4.44 |
| Carbohydrate | 3.74 - 14.23 | 7.44 | 6.74 - 10.89 | 8.59 | 6.50 - 10.14 | 8.14 |

 TABLE 2. Range and average of ash, crude fibre, lipids, protein and carbohydrate content (%) of Enteromorpha intestinalis, Sargassum salicifolium and Gelidium latifolium.

Ash

During the period of investigation, the amount of ash in the investigated algae varied sometimes slightly and sometimes considerably. The limits of variation in G. *latifolium* were 12.50 and 18.60%, in S. *salicifolium* 18.20 and 30.41% and in E. *intestinalis* 20.20 and 56.17%.

Pattern of changes (Fig. 4a) in ash content of G. latifolium was essentially similar to that of S. salicifolium. In both algae, the amounts of ash were at maxima in summer and at minima in autumn. On the other hand, ash content of E. intestinalis showed a maximum in winter and a minimum in spring (Fig. 4a).

Crude Fibre

Throughout the whole period of investigation, the crude fibre content of G. *latifolium* was consistently higher than that of E. *intestinalis* and S. *salicifolium* (Table 2 & Fig. 4b).



FIG. 3. Seasonal variations in contents of inorganic nitrogen and elements of surface seawater at Abu-Qir.



FIG. 4. Seasonal variations in ash, crude fibre, lipids, protein and carbohydrate content of *E. intestinalis, G. latifolium* and *S. salicifolium*.

From Fig. 4b, it is clear that the maximum concentrations of crude fibre in the investigated algae were recorded in summer. In *G. latifolium* and *S. salicifolium*, the crude fibre content showed the minimum in winter. In contrast, *E. intestinalis* showed a minimum in spring.

Lipids

Marked seasonal variations were observed for lipid content of E. intestinalis and S. salicifolium. Furthermore, the pattern of changes in lipid content of E. intestinalis was essentially similar to that of S. salicifolium. The highest values were recorded in summer while the lowest were obtained in autumn (Fig. 4c).

In G. latifolium, there was little variation from season to season, but in winter the highest amount was recorded (Fig. 4c).

Protein

Protein content of the investigated algae was characterized by remarkable seasonal variations (Fig. 4d). In *E. intestinalis* and *S. salicifolium*, the protein was high in winter and low in summer and spring respectively. *G. latifolium* exhibited the minimum in autumn and maximum in summer. *S. salicifolium* was distinctive for its high content of protein (Table 2).

Carbohydrates

Carbohydrate content of the three algae exhibited noticeable seasonal variations. All algae showed maxima in winter. The minimum values were observed in spring and summer (Fig. 4e). *E. intestinalis* generally had a lower carbohydrate content than *S. salicifolium* and *G. latifolium* (Table 2).

Discussion

A comparison of the fluctuations occurring in the environmental factors with those occurring in ash, crude, fibre, lipids, protein and carbohydrate content of *Enteromorpha intestinalis, Sargassum salicifolium* and *Gelidium latifolium* revealed that not all environmental factors examined acted similarly on these constituents. On the other hand, the effect of these factors was also species – specific.

As regards the influence of environmental factors on the ash content of the studied algae, it was noted that ash content of *G*. *latifolium* and *S*. *salicifolium* was reversible to pH values. Maxima and minima of ash content coincided exactly with minima and maxima respectively for pH of seawater.

For *E. intestinalis*, it was found that the ash content appear to be parallel to nitrate level, while it was reversible to concentration of potassium of seawater.

The rest of environmental factors did not show any direct influence on the ash content of the studied algae. In this connection it should be mentioned that Sway *et al.* (1991) reported that no correlation was found between ash content of *Caulerpa serrulata* and *Enteromorpha* sp. from Gulf of Aqaba and water temperature. The results also show that the lowest amounts of ash in *G. latifolium* and *S. salicifolium* were recorded when the maximum values of chlorides, organic carbon, silicate, ammonia, magnesium and sodium were determined. This would suggest that the high amounts of these factors may affect the ash content of the above two algae.

The data presented in this study show that the seasonal changes in crude fibre content of *S. salicifolium* and *G. latifolium* closely followed the same as those found for water temperature and total soluble salts, with high values in summer and low in winter. On the other hand, the crude fibre content of the above mentioned algae was also reversible to inorganic phosphorus and nitrite content of seawater.

As seen from the results, no direct relation could be deduced between the variations in the environmental factors and the crude fibre content of E. intestinalis.

The results further show that maximum amount of crude fibre of all algae was measured in summer concurrently with the maximum values of water temperature, sulphate and total soluble salts. This does not necessarily mean that the increase in content of the crude fibre is directly proportional to these factors. However, a similar result has been presented by El-Naggar (1980) who found that maximum content of crude fibre in *Ulva rigida* also accompanied the maximum content of total soluble salts and sulphate of seawater.

In the present work, it was noted that lipid content of *G. latifolium* varies inversely compared with water temperature and total soluble salts. On the other hand, *Gelidium* showed an exact parallelism between crude fibre content and concentration of inorganic phosphorus and nitrite of seawater.

Lipid content of *E. intestinalis* and *S. salicifolium* generally showed a close inverse relation with pH. Also, the data revealed that minimum amount of lipid in *E. intestinalis* and *S. salicifolium* was accompanied with the maxima of chloride, organic carbon, silicate, ammonia, magnesium and sodium.

On studying the effect of environmental factors on the protein content, it was found that fluctuation of protein content in E. *intestinalis* was reversible to water temperature and total soluble salts trends. Also, protein content of this alga proceeds parallel to inorganic phosphorus and nitrite content of seawater.

In *G. latifolium*, the protein content was reversible to pH value. Maximum and minimum of protein coincide exactly with minimum and maximum respectively for pH.

S. salicifolium shows an exact parallelism between protein content and nitrite content of seawater. Maximum and minimum of protein content coincided exactly with maximum and minimum for the nitrate content. In several other studies, the seasonal variations in protein-bound amino acids of brown algae paralleled those reported for seawater nitrate (Wheeler and Srivastava 1984, Wheeler *et al.*, 1984 and Rosell and Srivastava 1985). On the other hand, protein content of *S. salicifolium* was reversible to potassium content of seawater. Comparing carbohydrate content with the environmental conditions, it was found that carbohydrate content of *S. salicifolium* appear to be parallel with inorganic phosphorus and nitrite levels of seawater while it was reversible to total soluble salts.

E. intestinalis showed an exact parallelism between its carbohydrate content and nitrate amounts of seawater. Also, it was found that carbohydrate content of this alga was almost inversely proportional to potassium concentration of seawater.

With regard to G. latifolium, no direct relation between the carbohydrate content and the environmental factors could be deduced.

From the above discussion, it is concluded that there are numerous intricate factors affecting the major constituents of the studied algae. It is not correct to correlate these chemical constituents of such algae with a single factor. However, total soluble salts, potassium, nitrite, nitrate, inorganic phosphorus, pH and water temperature were the major controlling factors; other factors were merely contributory.

As it has been reported by several authors (Chapman and Craigie 1977, Chapman *et al.*, 1978 and Chapman and Lindley 1980) the amounts of available nitrogen may have a regulatory effect on the seaweed. Bird (1984) found that at increasing nitrate concentrations the protein: carbohydrate ratio of algal thalli increased. Sea-weed pigment contents were generally positively correlated with dissolved inorganic nitrogen (Duke *et al.*, 1986).

The present findings are similar to previous results by Haug and Larsen (1958a&b) which showed that the salt content of seawater exerts a pronounced influence on the chemical composition of *Ascophyllum nodosum*. Munda and Kremer (1977) found that the dry weight, ash, chloride and mannitol content of *Fucus vesiculosus* and *F. serratus* declined progressively with reduced salinity. However, Murthy and Radia (1978) noticed that salinity has no effect on composition of *Ulva lalctuca, Gelidella acerosa* and *Sargassum swartzii*.

Water temperature is among the environmental factors which affect fatty acid composition of marine algae (Khotimchenko and Svetoshev 1988). Changes in temperature are known to cause changes in composition and metabolism of lipids in few seaweeds (Pohl and Zurheide 1979 and Pettitt *et al.*, 1989).

As mentioned above, the chemical composition of the studied algae generally influenced by inorganic phosphorus content of seawater. Vollenweider (1968) points out that phosphorus is a major nutrient regulating growth in lakes. The chlorophyll content of *Codium decorticatum* increased with addition of P (Ramus 1983).

In view of the data presented in this investigation, the conclusion may be reached that the level of chemical constituents of marine algae depends both on the environmental conditions and on the species. Further experimental work is required to study the effect of each of the major controlling factors separately under controlled conditions.

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تأثـير العـوامل البيئيـة المختلفـة على المـكونات الكيميائيـة الكـبرى لبعض أنــواع الطحــالب البحريــة

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المستخلص . تضمن هذا البحث دراسة العلاقة بين المكونات الكيميائية الكبرى (الرماد - الألياف - الليبيدات - البروتين - الكربوهيدرات) لثلاثة أنواع من الطحالب البحرية وهي أنتيرومورفا انتستينالز (أخضر) وسرجاسم ساليسفوليم (بني) وجليديم لانيفوليم (أحم) وخمسة عشر نوعًا من العوامل البيئية .

دلت هذه الـدراسـة على أن هذه المكـونات الكيميائية قد تأثرت بعديد من العوامل البيئية . في نفس الوقت وجد أن تأثير هذه العوامل قد اختلف تبغًا لنوع الطحلب .

وقد تبين من نتائج هذه الدراسة أن كل من المحتوى الكلي للأملاح الذائبة والنيترات والنترات والبوتاسيوم والفوسفور غير العضوي ودرجة حرارة الماء والرقم الهيدروجيني هي أكثر هذه العوامل تأثيراً أما ماعداها فذات تأثير شبه محدود .