

To Prof. Gonyale
Best Wishes,
Charlie Yum.

Acclimation Responses to Salinity of Three Estuarine Red Algae from New Jersey

C. Yarish¹, P. Edwards^{1*} and S. Casey²

1536 ✓

¹Department of Botany, Rutgers, The State University; New Brunswick, New Jersey, USA and

²Department of Statistics, Rutgers, The State University; New Brunswick, New Jersey, USA

Abstract

The effects of salinity and acclimation time on the net photosynthetic responses of 3 estuarine red algae, *Bostrychia radicans* Mont., *Caloglossa leprieurii* (Mont.) J. Ag., and *Polysiphonia subtilissima* Mont., from Great Bay Estuary, New Jersey, USA, were investigated. The algae were cultured in a series of synthetic seawater media of 5, 15, 25 and 35‰ S for acclimation periods of 0, 2, 4, 8, and 16 days prior to determining their photosynthetic responses. All species were euryhaline, and demonstrated photosynthesis at all the above salinities. *B. radicans*, which was more common towards the mouth of the estuary, had a maximum photosynthetic rate at 25‰ S, whilst *C. leprieurii* and *P. subtilissima*, which were more common towards the head of the estuary, had photosynthetic maxima between 15 and 25‰, and at 15‰, respectively. The curves relating net photosynthesis to salinity were usually similar within a species at different acclimation periods, although statistically significant differences were sometimes noted. The acclimation periods producing maximal net photosynthesis were 0, 2 and 4 days for *B. radicans*, and 4 days for *C. leprieurii*, whilst for *P. subtilissima* there was no significant difference in response for any acclimation period over the range of salinities studied.

Introduction

Previous investigations on the effect of salinity on the net photosynthesis of benthic marine algae have revealed a variety of responses which suggest that the interactions are complex. The inability to define general trends stems in part from one or more of the following: a lack of sufficient replication which is necessary with plant material that may be heterogeneous; the ability of different species of seaweeds to thrive in a restricted (stenohaline species) or in a wide (euryhaline species) salinity range; differences in experimental techniques, particularly with regard to the acclimation of the plant material prior to running an experiment.

Fromageot (1923) reported that the highest rates of photosynthesis in several green algae occurred in full-strength seawater and that deviations in salinity in either direction caused a

decline in the rate of photosynthesis. This response suggests a relatively stenohaline group of algae. In contrast, the euryhaline species *Ulva lactuca*, *Fucus virsoides*, *Porphyra leucosticta* and *Wrangelia penicillata* from the Adriatic Sea, studied by Zavodnick (1975), were able to photosynthesize over a wide range of salinities, from 0 to 42‰ S.

However, there are reports in the literature that are more difficult to interpret. Legendre (1921) found that, in a reduced salinity of about 22‰ S, the photosynthetic rates of *Ulva lactuca* and *Fucus serratus* gradually increased to more than double the rate in full-strength seawater. Nellen (1966) studied salinity influences on photosynthesis in *Delesseria sanguinea* and *F. serratus* and varied both the salinity and exposure time. Some of her results indicate that changes in salinity caused a subsequent stimulation in performance, followed by a depression due to the detrimental effects of the salinity change, but that this was not always so.

Another potential source of variation in the response of algae to salinity

*Present address: Agricultural and Food Engineering Division, Asian Institute of Technology, P.O. Box 2754, Bangkok, Thailand.

changes is the period of time the plant material is acclimated to the salinity in question. Many investigators used no acclimation period (Ogata and Matsui, 1965; Gessner, 1969; Zavodnick, 1975), so that responses measured may result from shock on being transferred to a totally different regime; other workers apparently chose arbitrary acclimation times (Druehl, 1967, 5 days; Mathieson and Dawes, 1974, 4 days; Fralick and Mathieson, 1975, 4 days; Dawes et al., 1976, 3 days).

The present study was designed to study the net photosynthetic responses of laboratory cultured material of 3 estuarine red algae, *Bostrychia radicans* Mont., *Caloglossa leprieurii* (Mont.) J.Ag., and *Polysiphonia subtilissima* Mont., to variations in salinity as a function of acclimation periods ranging from 0 to 16 days. The observed photosynthetic responses to salinity variations are correlated with the horizontal or local distribution of the algae in their estuarine environment.

Materials and Methods

The unialgal cultures of benthic algae used in the study were isolated from the Great Bay Estuary, New Jersey, USA. The habitats, locations and dates of collection of each isolate are as follows: *Bostrychia radicans* Mont., upper eulittoral zone, wooden pilings, Marine Biology Station, 17 July, 1973; *Caloglossa leprieurii* (Mont.) J.Ag., mid-eulittoral zone, wooden pilings, Parkway Bridge, 13 July, 1973; *Polysiphonia subtilissima* Mont., lower eulittoral zone, wooden pilings, Parkway Bridge, 30 July, 1974. Stock cultures were maintained in von Stosch's enriched seawater media (as cited by Ott, 1966), at 25‰ S for *B. radicans* and at 15‰ S for *C. leprieurii* and *P. subtilissima*. The tanks containing the algae were maintained under an illumination of 2252 ± 269 lux at 25°C. Female plants of one clone each of *B. radicans* and *C. leprieurii* and a vegetative clone of *P. subtilissima* were grown from excised branch tips in 15 l plastic tanks containing 12 l of the enriched seawater media. Each tank was aerated with compressed air to avoid CO₂ depletion (Emerson and Green, 1934; Tseng and Sweeny, 1946), enhance growth (Colinvaux et al., 1965; Ogata et al., 1972) and provide circulation. The air was passed through two deionized water filters to minimize the chance of contamination. Cloned plants were used to reduce genetic variability and ensure that reproduction would not take place. Thus, all experiments for each species were conducted

on a single genotype in the vegetative state.

A preliminary experiment was carried out in which the net photosynthesis of each species was determined at salinities of 0, 5, 15, 25 and 35‰ without prior acclimation. A series of experiments was then conducted to investigate the effect of acclimation periods of 0, 2, 4, 8 and 16 days (in a Sherer Dual Jet Incubator, Model RT46 B-SE, Marshall, Michigan, USA) in a given salinity regime on the net photosynthesis of each isolate. A synthetic seawater medium (Ott, 1965) containing 5, 15, 25 and 35‰ total salt was used to insure the availability of CO₂ (Ogata and Matsui, 1965; Hammer, 1968). This medium also contained NaHCO₃. The concentration of carbonate buffers in the artificial seawater was the same in all salinities. The pH of the media was 7.5 to 8.0, regardless of salinity. Net photosynthesis was measured by the azide modification of the Winkler Method (Rand et al., 1976) and expressed as mg O₂ g⁻¹ dry weight h⁻¹. Each experiment consisted of 4 replicates with an incubation time of 3 h. The light intensity and temperature in the Sherer Incubators were held constant at 2252 ± 269 lux and $26^\circ\text{C} \pm 1^\circ\text{C}$. The data were analyzed by one- and two-way ANOVA's, and Tukey's multiple comparison of the means (= Tukey's *w*-procedure, Miller, 1966), and were tested for significant differences at the 5% level.

Results

The net photosynthesis of *Bostrychia radicans* previously cultured at and therefore acclimated to a salinity of 25‰, was uniform ($P < 0.05$) over a salinity range of 15 to 35‰ (Fig. 1). A decrease in net photosynthesis occurred at salinities less than 15‰, and no net photosynthesis was recorded at 0‰ S. During acclimation experiments, all plants died after 3.5 days immersion in a salinity of 0‰. The effects of acclimation time on net photosynthesis at different salinities are shown in Fig. 2, and a statistical analysis of the trends is presented in Table 1. There was no significant difference ($P < 0.05$) between acclimation regimes of 0, 2 and 4 days over a wide range of salinities. Acclimation periods of 8 and 16 days, however, led to statistically significant reductions in the net photosynthetic rate compared to the acclimation periods of 0, 2, and 4 days. The optimal rate of net photosynthesis in all acclimation regimes was at 25‰ S. There were marked declines in

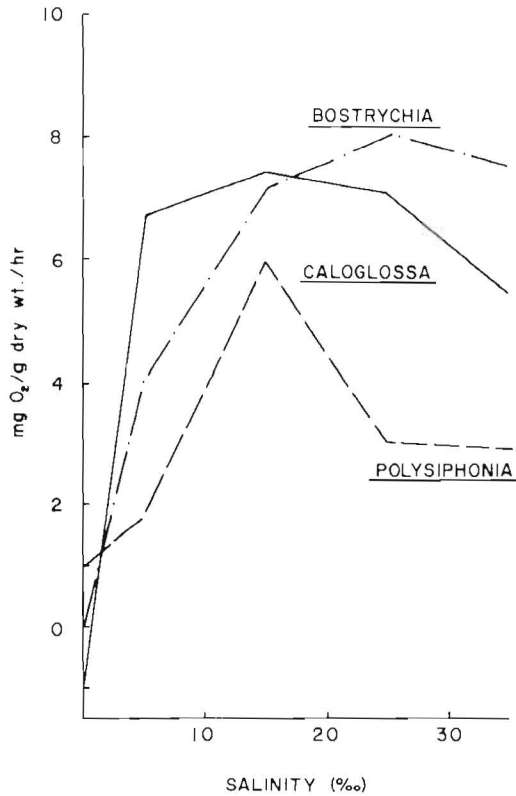


Fig. 1. *Bostrychia radicans* (Tukey's $w = 2.73$), *Caloglossa lepriurii* ($w = 1.55$), and *Polysiphonia subtilissima* ($w = 1.49$). Net photosynthetic rates after 3 h incubation at various salinities. Each point is based on 4 replicates. The algae had not been previously acclimated to the various salinities. *B. radicans* was cultured at 25‰ S, *C. lepriurii* and *P. subtilissima* at 15‰ S

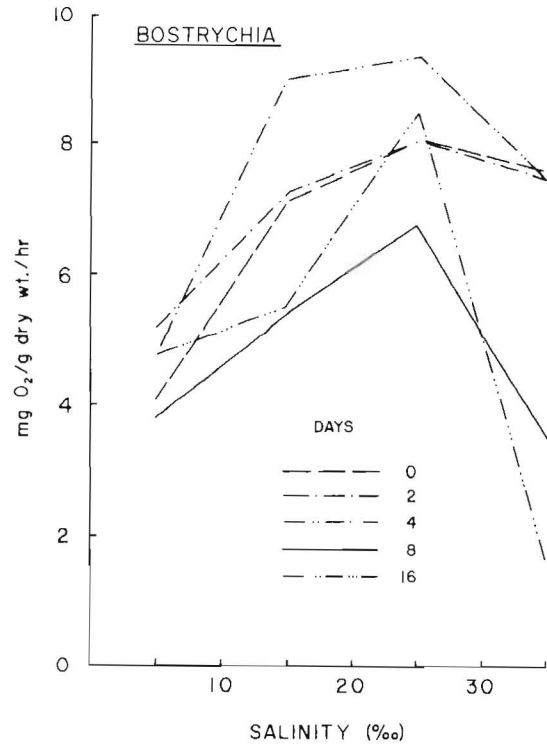


Fig. 2. *Bostrychia radicans*. Net photosynthetic rate with acclimation periods of 0, 2, 4, 8 and 16 days in salinities of 5, 15, 25 and 35‰ ($w = 2.75$). Each point is based on 4 replicates

Table 1. *Bostrychia radicans*, *Caloglossa lepriurii* and *Polysiphonia subtilissima*. Statistical summaries of net photosynthetic responses of clones to variations in salinity with different acclimation times. Analyses by Tukey's multiple comparison of means at the 5% level. Values underscored by same line are not significantly different from each other. Magnitude of photosynthetic responses are ranked from smallest to largest, left to right

Species	Treatment		Acclimation times (days)						
	Salinity (‰)								
<i>B. radicans</i>	5	35	15	25	<u>8</u>	<u>16</u>	<u>0</u>	<u>2</u>	<u>4</u>
<i>C. lepriurii</i>	<u>35</u>	<u>5</u>	<u>15</u>	<u>25</u>	<u>16</u>	<u>8</u>	<u>2</u>	<u>0</u>	<u>4</u>
<i>P. subtilissima</i>	<u>35</u>	<u>5</u>	<u>25</u>	<u>15</u>	<u>2</u>	<u>0</u>	<u>4</u>	<u>16</u>	<u>8</u>

net photosynthesis after 8 and 16 days acclimation at 35‰ S.

The net photosynthesis of *Caloglossa lepriurii* previously cultured at and therefore acclimated to a salinity of

15‰ was uniform ($P < 0.05$) over a salinity range of 5 to 25‰ (Fig. 1). A decrease in net photosynthesis occurred at salinities greater than 25‰, and no net photosynthesis was recorded at 0‰ S. The effects of acclimation time at different salinities are shown in Fig. 3, and a statistical analysis of the trends are given in Table 1. The optimal acclimation period over a broad range of salinities was 4 days (Table 1, $P < 0.05$). A significant decline in net photosynthesis occurred in both 8- and 16-day acclimation regimes at 5‰ S. The optimal rate of net photosynthesis in all acclimation regimes occurred between 15 and 25‰ S ($P < 0.05$).

The optimal rate of net photosynthesis of *Polysiphonia subtilissima* previously cultured at and therefore acclimated to a salinity of 15‰, occurred at 15‰ S (Fig. 1). A decline in net photosynthesis was apparent at salinities greater than or less than the optimum, and little photosynthesis was recorded at 0‰ S ($P < 0.05$). All plants died after 1.5 days immersion in a salinity of 0‰. The effects of acclimation time on net pho-

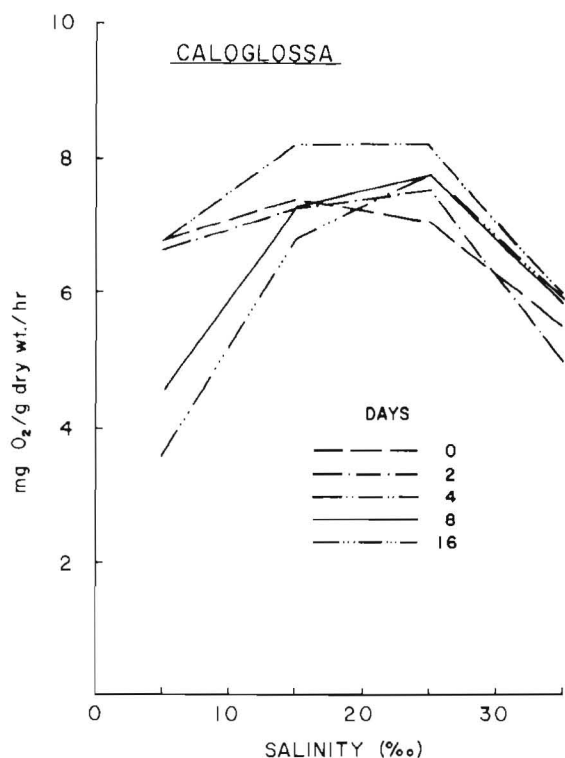


Fig. 3. *Caloglossa leprieurii*. Net photosynthetic rate with acclimation periods of 0, 2, 4, 8 and 16 days in salinities of 5, 15, 25 and 35‰ ($w = 1.48$). Each point is based on 4 replicates

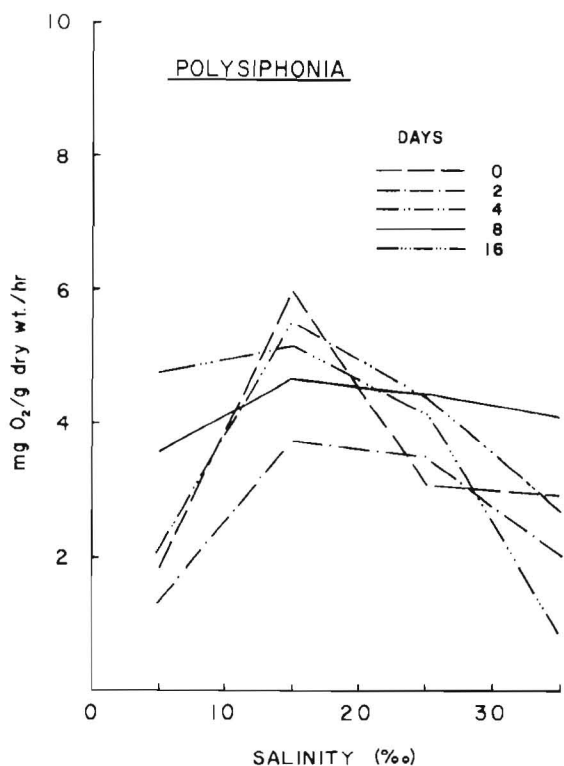


Fig. 4. *Polysiphonia subtilissima*. Net photosynthetic rate with acclimation periods of 0, 2, 4, 8 and 16 days in salinities of 5, 15, 25 and 35‰ ($w = 2.33$). Each point is based on 4 replicates

tosynthesis at different salinities are shown in Fig. 4, and a statistical analysis of the trends is presented in Table 1. There was no significant difference in response between acclimation times over the whole range of salinities tested ($P < 0.05$). The optimal rate of net photosynthesis in all acclimation regimes was at 15‰ S ($P < 0.05$). A distinct increase in net photosynthesis occurred at 35‰ S after 8 days acclimation.

Discussion

The patterns of net photosynthetic rates as a function of salinity variations differed for the 3 estuarine species of red algae studied, although all species were very euryhaline. *Bostrychia radicans* photosynthesized between 15 and 35‰ S (Figs. 1, 2) with a statistically significant maximum rate at 25‰ S (Fig. 2, Table 1). *Caloglossa leprieurii* had a relatively high photosynthetic rate at all salinities tested (Figs. 1, 3), with a statistically significant maximum rate between 15 and 25‰ S (Table 1). *Polysiphonia subtilissima* demonstrated a less broad response to salinity than the two preceding species, and had a distinct maximum rate at 15‰ S. All 3 species occurred throughout the estuary (Yarish and Edwards, 1976), with *B. radicans* being more abundant at the mouth (salinity range 23 to 31‰) and *C. leprieurii* and *P. subtilissima* being more common towards the head (salinity range at the Parkway Bridge 5 to 19‰). Thus, there is a general correlation between the horizontal or local distribution of these algae in the Great Bay Estuary System in New Jersey and their net photosynthetic responses to salinity variations in the laboratory.

None of the species occurred in fresh water, and this correlated with their limited survival time of 1.5 to 3.5 days in fresh water. More difficult to explain is the absence of the species from the relatively higher salinity regime of the open coast of New Jersey (monthly range for nearby Atlantic City on the open coast is 27 to 35‰ S, U.S. Coast and Geodetic Survey, 1965). All 3 species showed considerable photosynthetic activity at 35‰ S. However, *Bostrychia radicans* showed a large decline in activity at this salinity after 8- and 16-day acclimation periods, and *Polysiphonia subtilissima* after 16 days. Surprisingly however, the photosynthetic rate of *P. subtilissima* at 35‰ after an 8-day acclimation period, was the highest among all acclimation periods. The reduction of the photosynthetic rate could have been

due to the long acclimation period in artificial seawater, which may not be an adequate substitute for natural seawater. Certainly, the responses of all 3 species in enriched seawater media were similar to the responses of material acclimated in artificial media for up to 4 days, but plants were not acclimated in enriched seawater media.

However, the possibility cannot yet be ruled out that a salinity of 35‰ is too high for these species. This is also supported by Fralick and Mathieson (1975), who reported a decrease in the photosynthetic rate of *Polysiphonia subtilissima* at salinities greater than 30‰. On the other hand, Mathieson and Dawes (1975) have recorded *P. subtilissima* in Florida on the open coast in a salinity range of from 37 to 42‰. Such distribution differences might be due to ecotypic or physiological race variations.

There are conflicting reports in the literature on the effect of acclimating plants to a given salinity before determining their photosynthetic rate. Kjeldson (1967) and Ogata and Schramm (1971) reported that the rates of photosynthesis of non-acclimated plants in a given salinity were always higher than in plants acclimated to that salinity. Montfort (1931) stated that it may not be possible to establish a definite relationship between the photosynthetic rate of a given species and salinity, since the rate varies according to both salinity and exposure time, resulting in an almost unlimited number of curves. However, for each species in the present study, the curves relating net photosynthesis with salinity were similar for different acclimation periods, and most can be correlated with the horizontal distribution of the species in the estuary. Assuming that a maximal net photosynthetic rate implies that a species is fully acclimated, a 4-day acclimation period would appear to be the best one to use for the 3 species studied (Table 1). Therefore, it would be wise to do preliminary acclimation studies to identify a valid time period, since this study indicates that such an acclimation period would not be appropriate for all species.

Literature Cited

- Colinvaux, L.H., K.M. Wilbur and N. Watabe: Tropical marine algae: growth in laboratory culture. *J. Phycol.* 1, 69-78 (1965)
- Dawes, C.J., R. Moon and J. LaClaire: Photosynthetic responses of the red algae, *Hypnea musciformis* (Wulfen) Lamouroux (Gigartinales). *Bull. mar. Sci.* 26, 467-473 (1976)
- Druehl, L.D.: Distribution of two species of *Laminaria* as related to some environmental factors. *J. Phycol.* 3, 103-108 (1967)
- Emerson, R. and L. Green: Manometric measurements of photosynthesis in the marine alga *Gigartina*. *J. gen. Physiol.* 17, 817-843 (1934)
- Fralick, R.A. and A.C. Mathieson: Physiological ecology of four *Polysiphonia* species (Rhodophyta, Ceramiales). *Mar. Biol.* 29, 29-36 (1975)
- Fromageot, D.: Influence de la concentration en sels de l'eau de mer sur l'assimilation des algues vertes. *C. r. hebdom. Séanc. Acad. Sci., Paris* 177, 779-780 (1923)
- Gessner, F.: Photosynthesis and ion loss in the brown algae *Dictyopteris membranacea* and *Fucus virsoides*. *Mar. Biol.* 4, 349-351 (1969)
- Hammer, L.: Salzgehalt und Photosynthese bei marinen Pflanzen. *Mar. Biol.* 1, 185-190 (1968)
- Kjeldson, C.: Effects of variations in salinity and temperature on some estuarine macro-algae, 157 pp. Ph.D. thesis, Oregon State University 1967
- Legendre, R.: Influence de la salinité de l'eau de mer sur l'assimilation chlorophyllienne des algues. *C. r. Séanc. Soc. Biol.* 85, 222-224 (1921)
- Mathieson, A.C. and C.J. Dawes: Ecological studies of Floridian *Eucheuma* (Rhodophyta, Gigartinales). II. Photosynthesis and respiration. *Bull. mar. Sci.* 24, 274-285 (1974)
- Seasonal studies of Florida sublittoral marine algae. *Bull. mar. Sci.* 25, 46-65 (1975)
- Miller, R.: Simultaneous statistical inference, 272 pp. New York: McGraw Hill 1966
- Montfort, C.: Assimilation und Stoffgewinn der Meeresalgen bei Aussüßung und Rückversalzung. *Ber. dt. bot. Ges.* 49, 49-66 (1931)
- Nellen, U.R.: Über den Einfluß des Salzgehaltes auf die photosynthetische Leistung verschiedener Standortformen von *Delesseria sanguinea* und *Fucus serratus*. *Helgoländer wiss. Meeresunters.* 13, 288-313 (1966)
- Ogata, E. and T. Matsui: Photosynthesis in several marine plants of Japan as affected by salinity, drying and pH, with attention to their growth habitats. *Botanica mar.* 8, 199-217 (1965)
- and H. Nakamura: The life cycle of *Gracilaria verrucosa* (Rhodophyceae, Gigartinales) *in vitro*. *Phycologia* 11, 75-80 (1972)
- and W. Schramm: Some observations on the influence of salinity on growth and photosynthesis in *Porphyra umbilicalis*. *Mar. Biol.* 10, 70-76 (1971)
- Ott, F.D.: Synthetic media and techniques for the xenic cultivation of marine algae and flagellata. *Va J. Sci.* 16, 205-218 (1965)
- A selected listing of xenic algal cultures, 45 pp. Systematics-Ecology Program. Marine Biological Laboratory, Woods Hole, Mass. (mimeo): 1966 (Contr. Woods Hole oceanogr. Instn)
- Rand, M.C., A.E. Greenberg and M.J. Taras: Standard methods for the examination of water and wastewater, 14 ed. 1193 pp. Washington, D.C.: American Public Health Association 1976

- Tseng, C.K. and B.M. Sweeny: Physiological studies of *Gelidium cartilagineum*. I. Photosynthesis. With special reference to the carbon dioxide factor. *Am. J. Bot.* 33, 706-715 (1946)
- U.S. Coast and Geodetic Survey: Surface water temperature and salinity, Atlantic coast, North and South America, 88 pp. Washington, D.C.: U.S. Government Printing Office 1965. (Publs U.S. Cst geod. Surv. No. 31-1)
- Yarish, C. and P. Edwards: A field and cultural investigation of the seasonal and horizontal distribution of estuarine red algae of New Jersey, 124 pp. Ph.D. thesis, Rutgers, The State University 1976
- Zavodnick, N.: Effects of temperature and salinity variations on photosynthesis of some littoral seaweeds of the North Adriatic Sea. *Botanica mar.* 18, 245-250 (1975)
- Dr. Charles Yarish
Biological Science Group
University of Connecticut
Stamford Campus
Stamford, Connecticut 06903
USA

Date of final manuscript acceptance: December 1, 1978. Communicated by M.R. Tripp, Newark