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THE DAILY INTAKE AND DEGREE OF ABSORPTION OF THE SEA URCHIN PARACENTROTUS LIVIDUS FED UPON *CAULERPA TAXIFOLIA* (CHLOROPHYTA), *CYSTOSEIRA COMPRESSA* AND *HALOPTERIS SCOPARIA* (FUCOPHYCAE)

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The tropical green algua *Caulerpa taxifolia* (Vahl) C. Agardh was introduced into the Mediterranean sea in 1984 (MEINEZ and HESSE, 1991), where it forms very dense populations in the infralittoral zone, in particular between Nice and Menton (Alpes-Maritimes, France). *Caulerpa taxifolia* is toxic due to the production of some toxic terpenoids (GUERRIERO *et al.*, 1992 ; LEMEE *et al.*, 1993). The algas' toxicity changes greatly according to the season : in March-April, it is as its weakest and is most toxic from July to November (LEMEE *et al.*, 1993). During the hot season the sea urchin *Paracentrotus lividus* (Lamarck, 1816) notably avoids *Caulerpa taxifolia* ; on the other hand, during the cold season, the urchin is likely to feed upon the alga; however, the gonads of urchins fed upon *Caulerpa taxifolia* are found to be significantly less developped when compared with those of urchins fed upon a control alga (LEMEE *et al.*, 1994). The purpose of these experiments is to understand the reasons for these observations.

The experiments were carried out in aquariums, between the months of March and May 1994. The temperature of the water in the aquariums was constantely adjusted to correspond with the temperature of the sea in the region of the Alpes Maritimes. The algae offered to the urchins (*C. taxifolia*, *Cystoseira compressa* (Esper) Gerloff and Nizamuddin, *Halopteris scoparia* (Linnaeus) Sauvageau were gathered less than one week beforehand (except for experiment 2). The intake was measured by the daily weighing of the algae (measurements adjusted to take into account any growth of the algae). The degree of absorption was measured by calculating the difference between the intake and the faecal weight.

In all the experiments the urchins display phases of 1-3 days of feeding divided by phases of fasting lasting 1-2 days. These phases explain the significance of the standart deviations (Table 1). The intake of urchins fed upon *Caulerpa taxifolia* is significantely lower than those of urchins fed upon *Cystoseira compressa* and especially *Halopteris scoparia*, two algae considered to be moderately or strongly prefered, respectively by *Paracentrotus lividus*. The degree of absorption is found to be between 0 and 7% for urchins fed upon *Caulerpa taxifolia* as opposed to between 7 and 34% for urchins fed upon the two other algae. Futhermore, if *Caulerpa taxifolia* is freshly gathered (experiments 1, 3 and 4), all urchins feeding upon it are found to be dead within 14 to 18 days after the beginning of the experiment. On the other hand, there is no mortality for the urchins fed upon *Caulerpa taxifolia* conserved for longer than 15 days in an aquarium (experiment 2). However, the intake and the degree of absorption (this in particular) remain low.

Algae offered	Experiment N°	Dates	Daily Intake Mean	Daily Intake Standard deviation	degree of absorption %
Caulerpa taxifolia	1	March 25 - April 4	8	6	0
	2	April 18 - May 19	41	32	2
	3	April 10 - April 22	13	19	0
	4	May 6 - May 20	42	38	7
Cystoseira Compressa	5	March 25 - May 19	89	55	34
	6	April 18 - May 19	59	45	7
Halopteris scoparia	7	March 25 - May 19	134	96	23
	8	April 18 - May 19	121	77	13

Table 1 : Intake (in mg of dry weight/day/individual) and the degree of absorption (as a % of the mass ingested) of *Paracentrotus lividus*.

In the cold season, when *Caulerpa taxifolia* is at its least toxic, *Paracentrotus lividus* will feed upon the alga. However, the intake and the degree of absorption (this in particuler) are very low, which explains the observed mortalities, as well as the underdevelopement of the gonads reported by LEMEE *et al.* (1994). Furthermore, the conservation in an aquarium of *Caulerpa taxifolia* probably alters its chemical composition and hence this parameter must therefore be taken into account in the experimental protocols.

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SEASONAL EFFECTS OF *CAULERPA TAXIFOLIA* (VAHL.) C.AGARDH ON THE GROWTH OF *PHAEODACTYLUM TRICORNUTUM* BOHLIN

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Caulerpacean algae sinthetize many toxic secondary metabolites as a defense against grazers and epiphytes (PATTERSON *et al.*, 1984). Recently introduced populations of *Caulerpa taxifolia* (Vahl) C. Agardh in the Mediterranean sea also produce these substances, some of them (caulerpenyn) even in higher concentration than in the tropical ones (GUERRIERO *et al.*, 1992). This toxicity is liable to seasonal variations, as shown by toxicity experiments against mice, urchins and mammalian cells (LEMEE *et al.*, 1993). Since the mediterranean populations of *C. taxifolia* are extremely denses, an undesirable impact may be produced on both planktonic and benthic microalgae. As they are the first step in the food web and play an important ecological role in the sea, if microalgae population are affected by *C. taxifolia*, the whole ecosystem will be.

Our goal is to check whether *C. taxifolia* is toxic for some marine microalgae growing in laboratory culture conditions and to verify the existence of seasonal differences in the *C. taxifolia* effect on them. We will show the results obtained for *Phaeodactylum tricornutum* Bohlin a pennate diatom common in supralittoral rock pools, and extensively used in laboratory work and aquacultural systems to feed the invertebrate juveniles. *C. taxifolia* was collected at days 20/11/93, 28/2/94, 22/5/94 and 1/9/94 on the French coasts of Cap Martin at 9 m depth, and immediately transported to the laboratory in aerated opaque containers. Each toxicity test were started the day after. Unialgal cultures of *P. tricornutum* were supplied by Dr. Lubian, ICMA (CSIC), Cadiz. Cultures were maintained in artificial seawater (ADSA-Micro) enriched with f/2 Guillard's medium (GUILLARD & RYTHER, 1962) in a growth chamber set at 20°C on an alternating 12:12 LD cycle at 100 $\mu E.m^{-2}.s^{-1}$ cool-white fluorescent lighting. Subcultures were previously acclimated for a period of 2 weeks at the temperature at which the experiment was to be conducted (the same registred in the sea when *C. taxifolia* were collected: 15°C, 12°C, 17°C and 25°C respectively). Six culture vessels containing 200 ml sterile medium were inoculated to give initial population densities around 10^4 cells ml^{-1}. Simultaneously, two fragments consisting on both blades and stolons of *C. taxifolia* (2 g each) were added simultaneously to 4 of them. One millilitre aliquots from thoroughly mixed cultures were withdrawn daily for a period of 27 days (25 days for winter experiment), and preserved with a drop of formaldehyde. Cell counts were performed with a Coulter Multisizer-2.

	Autumn		Winter		Spring		Summer	
	Cont.	Caul.	Cont.	Caul.	Cont.	Caul.	Cont.	Caul.
Max. Growth Rate	0.84	0.29	0.72	0.57	1.75	0.56	0.58	0.1 5
Max. Cell Concentration	28	11	15	2.6	4.8	1.4	4.3	1.3

Table I. Growth rate (div./day) and maximum cell concentration (cells/ml x 10^5) for both control (Cont.) and test (Caul.) experiments.

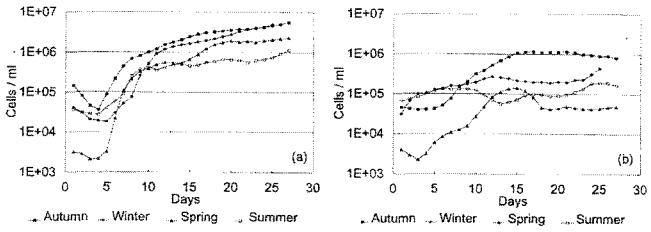

Fig. 1 Growth curves for control (a) and test cultures (b) in the four seasonal experiments.

Smoothed growth curves (fig. 1) showed a less growth in test cultures than in controls, in every season. Sligthly different profiles among control growth curves could be explained by temperature and initial cell concentration. Obviously, test cultures were also affected by these two factors, but differences among the control and test curves were too large for these factors to account for them. There were observed different behaviours in the growth of test cultures among the different seasons. A lag-phase was present in the autumn and spring test cultures but not in the other seasons. The slope and the lenght of the exponential growth phase changed in the different seasons, and so the number of cells at the end of the exponential growth changed too. When maximum growth rates and maximum cell concentrations were considered it appeared that, in all the seasons, they both were lower in test cultures than in controls (Table I). Maximum growth rate pointed to summer and spring test cultures as the most different to the controls (3.85 and 3.12 times lower) (Table I). Maximum cell concentration for spring and summer experiment were 3.54 and 3.22 times lower in test cultures than in controls (Table I) and although winter experiment showed the highest difference between test and control cultures (5.77 times lower) this result, due to a ciliate proliferation had to be discarded. The ciliate growth could be the cause of the inhibition of the diatom growth instead of the *C. taxifolia* effect. Moreover, in this season collected *C. taxifolia* was strongly epiphyted. That fact together with the ciliate growth might point out that the macroalga was less toxic (LEMEE *et al.*, 1993; DINI *et al.*, 1992).

Then, we can conclude that, in the experimental conditions described, control/test culture ratios of the maximum growth rate and maximum cell concentration at the end of the exponential phase were the best features to compare the effect of *C. taxifolia* in different seasons and that *P. tricornutum* is highly affected by *C. taxifolia*, specially in summmer and spring. Anyway, the extrapolation of these results to the natural environment is to be considered carefully since culture conditions in the laboratory differ from the environment where *C. taxifolia* developes.

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