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A TEST ON THE USE OF *CLADOPHORA PROLIFERA* (ROTH.) KUTZ. (CHLOROPHYTA, CLADOPHORALES) AS EFFECTIVE FERTILIZER FOR AGRICULTURAL USE

RIASSUNTO

Nel presente lavoro è stata testata l'azione delle polveri dell'alga verde *Cladophora prolifera* (Roth.) Kutzing come fertilizzante. La stessa alga è stata proposta quale organismo biorimediatore (estrattore di composti azotati) nel trattamento dei reflui derivanti dall'attività di acquacoltura.

L'esperimento è stato effettuato su piantine di pomodoro *Lycopersicon esculentum* (Miller), con due soluzioni a diversa concentrazione di polvere di *C. prolifera* (10% e 5%). Ambedue hanno dato un esito positivo, inducendo una crescita maggiore nelle piante trattate, anche se la soluzione al 5% ha dato i migliori risultati.

INTRODUCTION

Since many years, seaweeds have been used as sources of chemical and biological products. Most of the literature deals with red algae, originally cultivated in Japan, and now commonly used in the alimentary and pharmaceutical fields (FUJIWARA-ARASAKI *et al*., 1984; NISIZAWA *et al*., 1987). By contrast, brown and green algae resulted still poorly investigated from this point of view (ARIELI *et al*., 1993; JURKOVIC *et al*., 1995; SERFOR-ARMAH *et al*., 1999).

Beside their use as food, fertilizer and phycocolloids production, macroalgae find application in the removal of nutrients from effluent water of sewage industry, mariculture land-based fishponds and cages (MSUYA and NEORI, 2002; TROELL *et al*., 1997; 2003). The yearly biomass produced as a by-product of bioremediation can feasibly be removed from the system and utilized, resulting in both environmental and economical advantages.

Our paper focuses attention on the green macroalga *Cladophora prolifera* (Rothpletz) Kutzing (Clorophyta: Cladophorales), a species distributed in the eutrophic environments of the Mediterranean, as well as along the Atlantic coast from Spain to Ireland.

Cladophora prolifera was firstly studied in the Ionian Sea by PARENZAN (1970) who observed an abundant population in the Porto Cesareo area, estimating a biomass of 14,000,000 Q. Chemical investigation, revealing the alga content-rich in important mineral oligoelements, lead this author to propose its utilization as a food-stock. The protein content of this alga is very high (24,62%) (BONOTTO *et al.*, 1987), especially when compared to values obtained from other green algae utilized by the food industry (FLEURENCE, 1999).

Cladophora prolifera has been recently inserted in a large bioremediation project for Nitrogen reduction in a polycolture system of an area located in the Porto Cesareo Marine Protected Area (Ionian Sea) (GIANGRANDE *et al.*, 2003). The project, which is still in progress, also includes a study on the growth and production of the alga with relatively simple cultivation techniques. Moreover preliminary experiments indicated for this species a high ammonium uptake rate (unpublished data).

In this paper we quantitatively assessed the effect of pulverized algal biomass on the growth and production of tomatoes, to exemplify the utilization of its biomass as fertilizer.

MATERIAL AND METHODS

Cladophora prolifera was collected in Porto Cesareo using a handle net. Material was collected during April 2003. Samples were transported to the laboratory where epiphytes were removed by successive washing with distilled seawater and rapid exposure to fresh water.

Cladophora prolifera was plugged with blotting paper; excess water was evaporated in a stove at 60° for 72 hours. This preparation was subsequently reduced in powder by means of a blender and preserved in glass jars. The fertilizer mixture for the tomatoes was prepared mixing the powder with water. Two different concentrations 5% and 10%, were prepared and tested.

In order to test the efficiency of the fertilizer, 6 compartments for seeds were prepared in a field of 6*4 m in size. Two rows of 9 tomato plants were put in each compartment. Single compartments and rows of plants were maintained separate to avoid the margin effect. Two compartments were left without fertilizer to act as controls. Two compartments were treated with the 5% solution of the fertilizer and the other two were treated with a 10% solution. In both treatments fertilizer

(200 cc of solution) was added to every plant 15 days after the transplant. Plants were transplanted on June 15, 2003 and fertilizer was added on June 30 in the two treatments. The first fruits appeared at the beginning of August. Afterwards, fruits were collected at intervals of about one week until September 15, when the last fruits were produced. Differences between controls and different treatments were measured in term of plant survival from the transplant (June 1) to the appearing of fruits (August 6) and in terms of production from August to September. Survival was investigated two times. The first one a month after the enrichment and a second one prior to harvesting the fruits. Data were analyzed with ANOVA Single factor. The tomato species utilized, *Lycopersicon esculetum* (Miller), is a local Apulia variety. This variety is highly resistant to pathogens and grows well in arid environments. The round fruits have a diameter from 10 to 14 cm, the colour is always reddish-orange and the calyx is present when harvested.

RESULTS

The maximum rate of fruit biomass was observed at the middle of the harvest (August 25th) both in the controls and treatments, with the largest disparity between the 5% treatments and the controls (Figure 1). Comparisons between controls and treatments, however, were not statistically supported (P>0.05) with the exception of

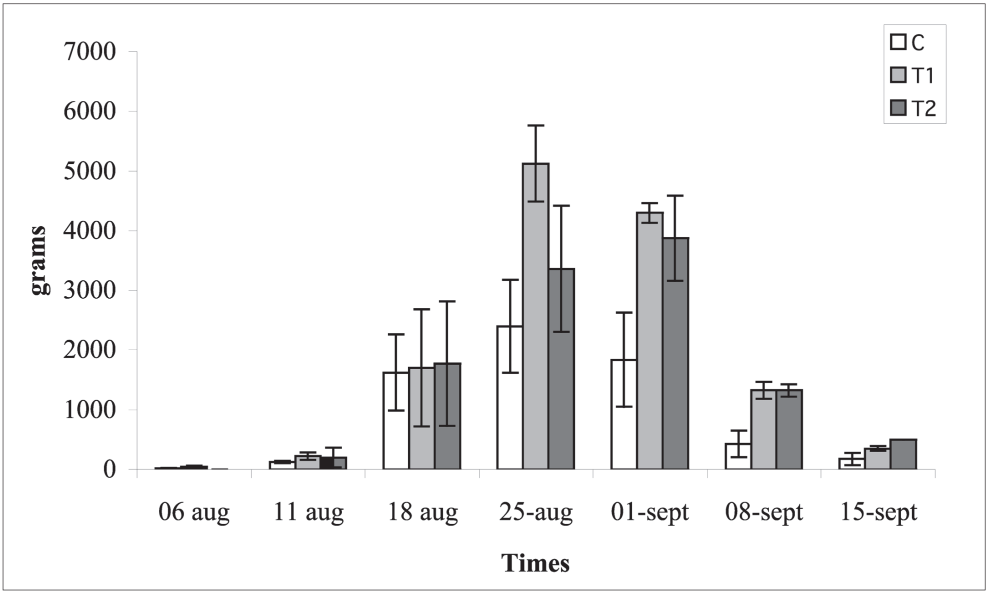

Fig 1 - Comparison of tomato production in time, among control and treatments, with harvesting maintained separated (C= control; T1 = 5% treatment; T2 = 10% treatment).

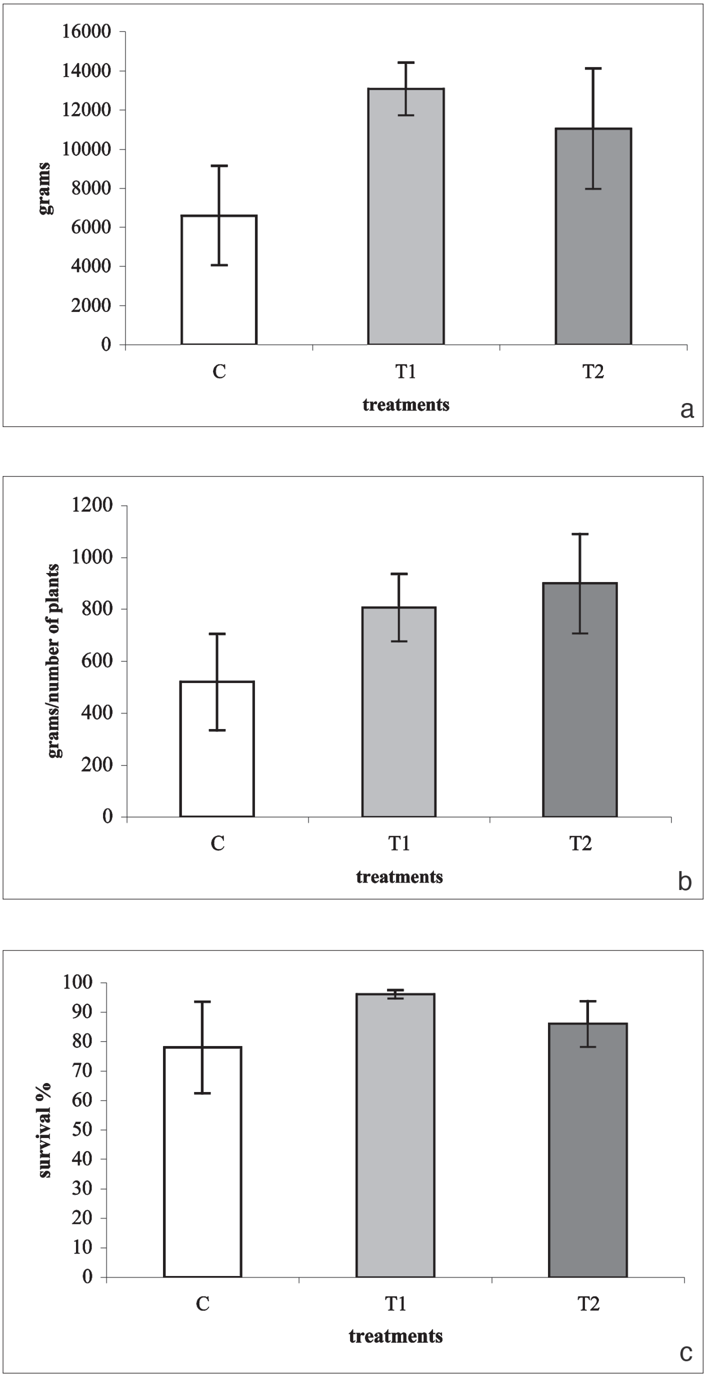

Fig. 2 - Field experiment: comparison among control and treatments: a) total production; b) mean production for plant; c) percentage of plant survival (C = control; T1 = 5% treatment; T2 = 10% treatment).

the last two records (Sept 8th and Sept 15th). As a whole, the weight of tomato fruits (total production) was higher in the 5% treatments relative to the controls (14,000 grams *vs.* 6,000), but also the 10% treatments compared to the controls showed a higher production (11,000 grams *vs.* 6,000). The total production appeared therefore higher at 5% treatment than at 10% treatment (Figure 2a). However, when the mean production (measured as production per plant) is examined, the 10% treatment showed the best result (Figure 2b). This was due to the difference in survival rate among treatments. The maximum of survival (94%) was observed in the 5% treatment, followed by the 10 % treatment with 86% of survival, while a survival of only 78% was found in non-treated population (Figure 2c).

Differences between controls and treatments were statistically significant only in the comparison between controls and 5% treatments ($P < 0.05$).

DISCUSSIONS AND CONCLUSION

The field experiment on the utilization of the seaweed biomass of *C. prolifera* as fertilizer has showed a significantly higher production and survival in treated plant. The higher production was obtained with a 5% solution. However, even though the total production was higher at 5% treatments compared to the 10% treatments, the production per plant was greater at 10% treatment. Fewer plants in fact survived when treated with the 10% solution, as a result the total production was higher at 5% treatments. It is interesting to note that plant survival is always higher in both respect to the controls. However, the variability between replicates was always very high, both in treatments and in controls, reflecting the existence of a high natural variability in plant survival and production. For this reason data are not always statistically supported, and only total production at 5% resulted significantly different from the control.

Although probably field experiment on the influence of algal powder on plant survival and production must be repeated, considering more replicates as possible, the results obtained from the present paper appear important for a future utilization of the biomass of this alga as natural fertilizer, also considering that the biomass can be a by-product from wastewater treatment.

As already mentioned, ammonium uptake rate of *C. prolifera* is quite high and it is possible that it is accumulated within thalli (unpublished data). The storage of accumulated ammonium in the thalli of some cultured macroalgae, which were observed to remove more nitrogen than they need, was already hypothesized by MSUYA and NEORI (2002). Present data seems to confirm that *C. prolifera* is an interesting ammonium store. However, nothing is known about the ammonium accumulation in *C. prolifera* thalli. In a previous study Schramm and Booth (1981), showed the capacity of *C. prolifera* to accumulate phosphorus. Further studies are in progress about analyses on chemical composition of the thalli also to search for substances for future utilization of this macroalga in food industry.

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