The effects of *Caulerpa taxifolia* on invertebrate abundance in Agua Hedionda Lagoon, California.

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Abstract Sea beds composed of eel grass (*Zostera marina), Caulerpa taxifolia* and both vegetation types were sampled at a lagoon in San Diego County at the first known invasion site of the Caribbean algae (*C. taxifolia)* in the United States. The hand-collected samples captured organisms from the surrouding water column, top sediment layer and vegetative matter. The abundance of Isopoda, Amphipoda and Spirorbidae populations were quantified. Weight adjusted samples were analyzed by means of the non-parametric Mann-Whitney U-test. The most significant decrease ($p<0.05$) in abundance of all three taxa was between the eel grass and Caulerpa samples. The effects of *C. taxifolia* invasions, and the ramifications on the benthic ecology of nearshore lagoons, could be devastating to the basal levels of the food chain.

Introduction

The challenges of dealing with invasive species have become of global concern with the relative ease of transcontinental movement (Ricciardi *et al,* 2000). Marine invasive organisms are more prevalent than their terrestrial counterparts due in part to the unwitting transmission on the hulls and in the ballast water of ships. The accidental introduction of *C. taxifolia*, an algae native to the Caribbean Sea, into the Mediterranean Sea in 1984 occurred when a small piece of algal material exited out of a flow-through aquarium in Monaco (Meinesz and Hesse, 1991). This exotic alga now covers hundreds of hectares of the Mediterranean seafloor, with patches reported up to hundreds of kilometers from original release point (Joussen *et al*., 1998).

The spread of *C. taxifolia* is due to an initial lack of eradication efforts and the amazing adaptability of this species to new environmental conditions, including colder water temperatures (Chrisholm *et al.*, 2000). Other characteristics of the algae that hinder containment efforts are the use of fragmentation as a method of dispersal (Ceccherelli and Cinelli, 1999; Smith and Walters, 1999) and the lack of suitable biocontrol organisms (Lemee *et al.*, 1996; Coquillard *et al.*, 2000; Zuljevic *et al.*, 2001; Thibaut *et al.*, 2001). The absence of biocontrol organisms is due to the production of caulerpenyne by *C. taxifolia*. Caulerpenyne is a naturally occurring neurotoxin that severely retards grazers (Pedrotti *et al.*, 1996; Bitou *et al.*, 1999; Ricci *et al.*, 1999; Pedrotti and Lemee, 1999). Studies correlating the detrimental effects of *C. taxifolia* on invertebrates have focused on physiological impacts caused by the neurotoxin caulerpenyne (Mozzachiodi *et al*, 2001). The majority of research focusing on the non-caulerpenyne affects of *C. taxifolia* on fauna has been conducted on fish. These studies have described qualitative and quantitative changes in fish communities (Relini *et al.*, 1998; Relini *et al.*, 2000) and specific physiological changes in some fish (Uchimura *et al.*, 1999). *C. taxifolia* is also known to cause a reduction in density and abundance of native sea-grasses in the Mediterranean Sea (De Villele and Verlaque, 1995; Ceccherelli and Cinelli, 1997; Ceccherelli and Cinelli, 1998).

C. taxifolia was first discovered in the United States in June 2000 in Agua Hedionda Lagoon, California, and to date there have been no published studies conducted on the new infestation. Agua Hedionda is an ideal study site not only because it allows observations of a preliminary infestation of *C. taxifolia,* but also the lagoon has a pre-established baseline for native populations (MEC, 1995) which may be useful in identifying any opportunistic species. Study of this organism will be important on an ecological level if eradication efforts in the previously infested Huntington Harbor, CA and Agua Hedionda Lagoon are not successful. A decline in certain "keystone" species, including amphipods, may result in an eventual decline in all species dependent upon these organisms for food with ramifications extending up the food chain.

The objective of this study is to determine the effects of *C. taxifolia* upon the abundance of marine invertebrates by comparing population differences in infested areas with *C. taxifolia* with those colonized only by native sea grasses.

Methods

Description of study subjects Three taxa were chosen for analysis: Amphipoda, Isopoda and Spirobidae. Amphipods and isopods were chosen because they are two of the most dominate invertebrate phyla represented in Agua Hedionda Lagoon according to a 1995 technical report (MEC, 1995) and form a necessary trophic level near the bottom of the food chain (Borga *et al*, 2001). The calcareous tube-forming worms, Spirobids, were chosen to represent the effects of *C. taxifolia* on organisms that use eel grass as a primary substrate upon which they live. Infaunal and epifaunal organisms are reasonable choices as bioindicators owing to their close association with the flora in addition to their relative immobility (Lafont, et al., 2001; Nerbonne and Vondracek, 2001).

Data Collection To assess changes in invertebrate abundance caused by the invasion of *C. taxifolia*, three cover types were chosen from within Agua Hedionda Lagoon, CA. The three types of cover are 100% *C. taxifolia*, a roughly 50-50 mix of *C. taxifolia* and eel grass, and 100% eel grass. All sample sites are located in the lagoon on the eastern side of Interstate 5. Sampled sites were within a $150m^2$ area of the lagoon and all samples were roughly equidistant from each other. The small alcove in which the sampling took place ranges from shallows (< 2 m) to depths of 8 m.

During the month of August 2000, 24 one-gallon samples were removed from the alcove. The samples were collected via use of a SCUBA device to diminish production of currents that could fragment and consequently spread the *C. taxifolia*. One-gallon plastic jars were used to collect the samples at an average depth of 4 meters below the surface of the water. The jars were dug into the sediment deep enough to obtain most of the rhizome mass anchoring the *C. taxifolia* in the Caulerpa and mix samples. The samples contained all of the vegetative material from each bunch of eel grass or *C. taxifolia*. Samples were immediately transported to shore, drained of water and filled with formalin. Proper storage of samples should be in 70% isopropyl alcohol. Due to financial and time constraints, samples were stored in formalin for approximately one year.

As each of the 24 jars was sorted, the formalin was filtered through a ½ mm. screen. The jar was filled with tap water and left to soak for one hour. The jar was again filtered through a ½ mm. screen to prevent loss of organisms but allowing sediments to filter out. The contents of the jar were emptied and washed to remove all sediment matter. A standard compound microscope (20x) was used to identify and quantify organisms in each jar. After classification, the plant matter and organisms were placed in glass jars in 70% isopropyl alcohol. The first three samples were analyzed during the summer of 2001. The remaining 21 samples were analyzed between February and April 2002.

A one-tailed Mann-Whitney U-test was used to compare the number organisms of each taxa in each of the eel grass, mix and *C. taxifolia* samples. For each taxon, p-values were multiplied by the number of comparisons made (n=3).

Results

Depicted in Figure 1 is the average number of organisms of each taxa divided by the weight of sample, without sediment, in grams. The Mann-Whitney U-test indicated that the strongest correlation denoting a significant difference in abundance of all three taxa, amphipods (p=0.0012), isopods (p=0.0012) and spirobids (p=0.0161) occurred between the eel grass and Caulerpa samples. Isopods were the only taxa that displayed significant differences in abundance in each of the three sample comparisons. P-values between mix and eel grass, and Caulerpa and mix samples were 0.0131 and 0.0312, respectively. In addition, amphipod (p=0.0095) and spirobid (p=0.0410) abundance was significantly lower when comparing mix and Caulerpa samples. All p-values are displayed in Table 1 with significant values in bold.

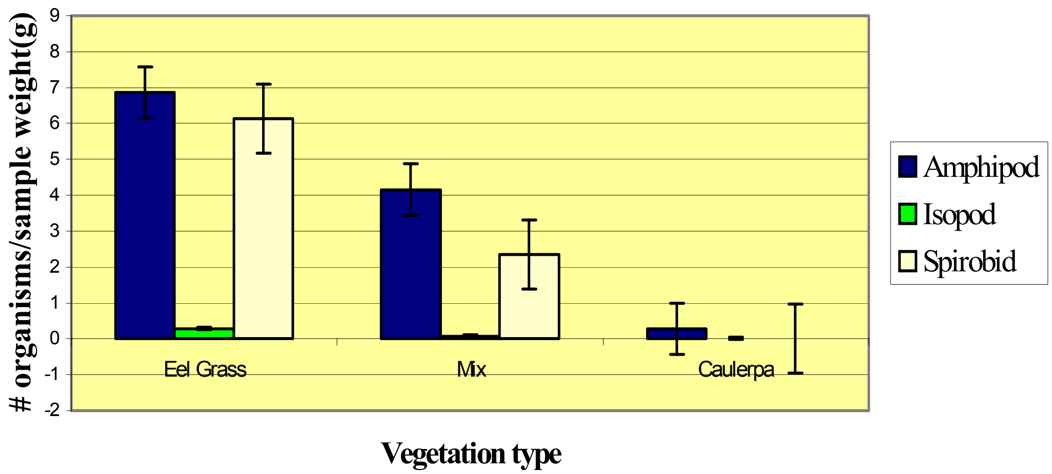

Figure 1. Mean values of each taxa in each vegetative type with average standard errors for each taxa

	Caulerpa v Mix	Mix v Eel	Eel v Caulerpa
Amphipoda	**0.0095**	0.1394	**0.0012**
Isopoda	**0.0312**	**0.0131**	**0.0012**
Spirorbidae	**0.0410**	0.4094	**0.0161**

Table 1. P values for each taxa between each comparison of vegetative type

Discussion

The significant decline in abundance of amphipods and isopods between the *C. taxifolia* and both other vegetation types would have ramifications along the food web in the lagoon. As previously noted these are two of the most abundant taxa in Agua Hedionda lagoon (MEC, 1995) and a decline in these may cause eventual decreases in abundance and possibly richness up the trophic levels. Sanchez-Moyano (2001) found that *Caulerpa prolifera* biomass sometimes became so great as to create anoxic conditions and physical impedements to invertebrate colonization. The Amphipod and isopod abundance declines observed in this study may be due to similar conditions created in Agua Hedionda lagoon by *Caulerpa taxifolia.*

One factor that may have resulted in nonsignificant findings for spirobid abundance was the age of the eel grass sampled. Some of the eel grass was young enough that spirobids had not yet colonized the stalks. It is interesting to note that no spirobids were found on any of the stalks of *C. taxifolia* in the strictly Caulerpa samples. However, three of the mix samples contained small numbers of spirobids on the stalks of the *C. taxifolia*. It is not known if the worms can eventually adapt to living solely on the *C. taxifolia* or if the caulerpenyne, the naturally occurring toxin, has an as yet unknown toxic effect on these organisms. The fact a few spirobids appear in the samples may mean that it is not toxic.

The appearance of the amphipod Leptostraca only in the Caulerpa seems to indicate it is an opportunistic species. More studies need to be conducted to confirm if a strong relationship exists between *C. taxifolia* and Leptostraca abundance. Clearly more avenues of research need to be pursued regarding potential opportunistic species and possible consequences of declines in invertebrate abundance to higher trophic levels. Although diversity of organisms in the three vegetation types was not the main focus of this study there was an observable decline in the richness between the eel grass and Caulerpa samples.

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