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Ecological Study on Community of Exotic Invasive Seaweed *Caulerpa prolifera* in Suez Canal and its Associated Macro Invertebrates

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Abstract: *Caulerpa prolifera* (Forsskal) Lamouroux, a green alga, widespread in tropical and subtropical seas is now invading species to the Suez Canal during last recent years after 2000; it is widely spread, colonizing its western sandy shore at shallow waters of 1-2 m depth. It has the potential to supplant native vegetation, thereby altering the structure and function of the subtidal marine landscape, supplant seagrass *H. stipulacea*. According to the present study, based on biometric parameters, the frequency of occurrence, abundance and density analyses, the seaweed *C. prolifera* is more frequent, abundant and dense in Suez Canal than the seagrass *H. stipulacea*, which is very rare. Instead *C. prolifera* forming extended dense meadows with percentage cover nearly 100% m^{-2} at many sites. This mainly happened; due to the competitive success of *C. prolifera* which seems to be related to its big size, high density, rapid growth, high efficiency in dim light conditions, high tolerance to severe nutrient limitation and salinity and temperature fluctuations and to the production of toxic secondary metabolites. The presence of these toxic secondary metabolites explains why *C. prolifera* is avoided by many of macro invertebrates as a habitat or feeding grounds.

Key words: Lake Timsah, Bitter lakes, Suez Canal, *Caulerpa prolifera*, seagrasses, macroinvertebrates

INTRODUCTION

The Suez Canal extends 172 km from Port Said in the north to the Suez in the South. About 80 km south of Port said it passes through Lake Timsah, some 5 km long and then after a further 15 km, through the 25 km of the Bitter lakes. The Canal and these lakes comprise the area of this study. These lakes contain nearly 90% of the water in the system and have predominantly sedimentary substrate (Por, 1978) and consequently support most of the fishing grounds. The hydrographic regime in these lakes is stable now with salinity of 43-47‰ at Bitter Lakes and 43‰ at Lake Timsah (Gab-Alla *et al*., 1990) with some freshwater input and agriculture run off which could lower the salinity at some sites near the western shore of both lakes (Gab-Alla *et al*., 1990).

Invasive species of the algal genus *Caulerpa* have garnered much attention in recent years (Sanchiz *et al*., 2001; Sanchez-Moyano *et al*., 2001) as they have the potential to supplant native vegetation, thereby altering the structure and function of the subtidal marine landscape. *Caulerpa prolifera* (Forsskal) Lamouroux is a green alga, widespread species in tropical and subtropical seas. It is a relative of the Mediterranean invasives of *Caulerpa taxifolia* and *Caulerpa racemosa* (Meinesz and Hesse, 1991; Meinesz, 1992; Meinesz *et al*., 1993a; Vaugelas *et al*., 1994) and commonly occurring, native, rhizophytic algal species along the Mediterranean (Taplin *et al*., 2005). Suez Canal began to be colonized by this seaweed (*Caulerpa prolifera*) as exotic invasive species since few years ago, after 2000 (Personal observation), where this species was not recorded before in the Suez Canal (El Manawy, 1987, 1992, 2000). But, last years, the progression of *C. prolifera* has been very rapid, expanding to cover most sandy substrate of Suez Canal, nowadays. This species has a record of stress on marine habitats (Sanchez *et al*., 2001; Sanchez-Moyano *et al*., 2001), with a great impact on different species and communities of algae, seagrasses, marine invertebrates and fishes. The presence of toxic secondary metabolites explains why *C. prolifera* is avoided by other marine biota.

In this study, we conducted a field survey to investigate the distribution of *C. prolifera* versus the seagrass *Halophila stipulacea* at different sites along Suez Canal to provide a potential explanation for pattern observed for the *C. prolifera* along Suez Canal according to marine environmental conditions related to its distribution. Also, to assess the associated diversity and the ecology of different associated macroinvertebrate fauna.

MATERIALS AND METHODS

Sampling was conducted at fourteen sites during April 2004 along the western coast of Suez Canal (Fig. 1).

Pilot survey showed that the seaweed *C. prolifera* was the most spreading flora on the sandy areas of Suez Canal.

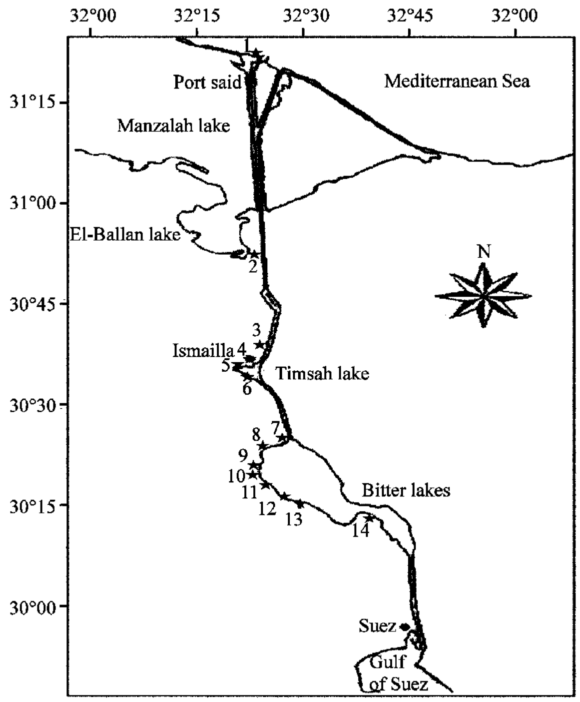

Fig.1: The sampling sites along the Suez Canal

At each sampling site, temperature (glass mercury thermometer), salinity (refractometer), water's depth were recorded. Quantitative sampling of the vegetation as well as, semi-quantitative visual observations of the epiphytes and macro-invertebrates of *C. prolifera* vegetation was carried out.

The abundance of vegetation was quantified by three methods: 1) total biomass in grams dry weight or ash free dry weight per square meter (gdwm^{-2}) 2) percentage cover (%m^{-2}) and 3) a modified Braun-Blanquet Cover-abundance Scale (Braun-Blanquet, 1965).

Vegetation density and biomass: Samples of seaweed or seagrass were obtained at each site by using 0.25 m^2 quadrat. To optimize the sampling process, four randomized samples were taken at each site (Kershow, 1980). Each sample consisted of all plant material with its roots and rhizomes to a depth of 15-20 cm in the substratum. After the quadrat was placed on the sea bottom, a knife was used to cut around its inside edge. This ensured that attached plant material outside the quadrat was not taken in the sample. Each quadrat was placed in a bag (5 mm mesh) and rinsed free of sediment. Macroscopic epiphytes (if present) were carefully scraped from the leaves.

In the laboratory, the plant material was rinsed in 5% phosphoric acid, to digest adhering carbon materials, then placed in an oven at 60°C and dried to a constant weight, then burned at 600°C in muffle ferns for determination of ash-free dry weight.

Braun-blanquet cover-abundance scale: Aach site, a minimum of four 1 m^2 (*H. stipulacea* or *C. prolifera*) quadrats were arbitrarily laid on the bottom. The seagrass or the seaweed occurring within the quadrats were assigned a cover-abundance scale value: 0.1-solitary, with small cover; 0.5-few, with small cover; 1-numerous, but less than 5% cover; 2-numerous, with 5-25% cover; 3-numerous, with 25-50% cover; 4-numerous, with 50-75% cover; 5-numerous, with more than 75% cover. Frequency of occurrence, abundance and density of the seagrass and seaweed were calculated by the following three formulae:

Frequency = No. of occupied quads/total No. of quads.

Abundance = Sum of Braun-Blanquet scale values/No. of occupied quads.

Density = Sum of Braun-Blanquet scale values/total No. of quads.

Benthic invertebrates: To investigate benthic communities at studied sites, belt transects (1×10 m) was established. The survey technique was done by observing and recording the distribution and the ecological status of different species (absent, rare, common) along transect.

Statistical analysis: Data of biometric parameters of the seaweed *C. prolifera* (percentage cover and biomass) and seagrass *H. stipulacea (*percentage cover, shoot number and biomass) at different sites were statistically tested by analysis of variance (One-Way ANOVA). Mean values were considered significant when $p<0.05$.

RESULTS

Environmental conditions: In the water column of Suez Canal, the salinity ranged from 10 to 44% and temperature from 23 to 26°C at different sites (Table 1). Sites with low salinity have usually access to fresh water input e.g., sites 12 (10%) and 13 (11%), or agriculture run off e.g., sites 5 (26%), 6 (32%) and 14 (37‰).The bay bottom is mainly sedimentary, with fine sand at uncovered sites or very fine sand at *Caulerpa* or seagrass sites except the second site which is rocky shore (Table 2). The depth at different sites is ranged from 1 to 2 m. The percentage of total organic carbon was high (6.94; 8.18%) at sites 7 and 11, while it was low (1.14, 1.18%) at sites 6 and 12, respectively.

Table 1: Status, water depth, temperature and salinity of different sampling sites along Suez Canal

Site	Position	Status	Depth (m)	Temp. (°C)	Salinity (‰)
1 Wave Barrier	31° 13` 20" N 32° 21` 70" E	Turbid marine water	1.5	24±2	39±1
2 Quantra	30° 51` 14" N 32° 18` 54" E	Clear marine water	1.5	24±1	42±2
3 El Ersal	30° 36` 47" N 32° 19` 14" E	Clear marine water	1.5	23±1	41±1
4 El Danva	30° 33` 50" N 32° 18` 12" E	Agriculture run off	2	25±1	38±1
5 El Faransawy	30° 33` 02" N 32° 18` 13" E	Agriculture run off	1.5	25±1	26±2
6 El Taaawen	30° 33` 04" N 32° 18` 14" E	Agriculture run off	1.5	26±2	32±2
7 El Deversoir	30° 24` 93" N 32° 21` 28" E	Agriculture run off	1	24±1	41±2
8 Abo Sultan	30° 22` 88" N 32° 18` 40" E	Tourist village	1.5	24±2	42±1
9 Fayed	30° 19` 29" N 32° 19` 14" E	Tourist village	1.5	25±1	43±1
10 El Sasater	30° 17` 40" N 32° 40` 46" E	Tourist village	2	24±2	44±1
11 Fanara	30° 17` 00" N 32° 18` 12" E	Tourist village	1.5	25±2	43±1
12 Kasfareet	30° 16` 54" N 32° 23` 23" E	Fresh water input	1.5	24±1	10±2
13 Rommana	30° 16` 00" N 32° 26` 82" E	Fresh water input	1.5	24±2	11±3
14 Kabreet	30° 15` 11" N 32° 28` 28" E	Agriculture run off	1	25±2	37±1

Table 2: Percentage of grain size (mm) and total organic carbon (% dry weight) of the bottom sediments of different sampling stations at Suez Canal

Station No.	Pebbles >2 mm	Very coarse sand 2-1 mm	Coarse sand 1-0.5 mm	Medium sand 0.5-0.25 mm	Fine sand 0.25 -0.125 mm	Very fine sand 0.125-0.063 mm	Silt and clay <0.063 mm	% TOC
1*	0.70	1.60	1.90	04.30	36.40	44.70	10.40	03.20
2**	-	-	-	-	-	-	-	-
3	1.69	01.96	03.23	06.63	40.35	43.20	04.61	02.49
4*	0.40	00.80	01.40	03.80	51.00	39.70	02.90	02.40
5*	1.50	02.60	04.60	06.30	35.40	17.60	32.00	05.14
6*	8.60	11.40	15.90	09.20	29.50	22.80	02.70	01.14
7	2.85	04.83	06.30	19.77	23.58	27.32	14.82	06.94
8	1.25	03.84	04.41	20.09	19.74	36.55	13.42	05.57
9	0.50	02.32	04.89	09.40	26.89	53.44	02.04	05.24
10	3.99	05.30	08.31	17.09	27.44	28.84	08.66	06.15
11	2.08	02.92	03.19	28.78	23.65	28.80	06.10	08.18
12*	6.50	06.00	05.00	04.00	49.60	22.70	06.10	01.48
13*	4.80	05.30	07.60	26.00	41.60	11.50	03.20	02.50
14	4.96	03.78	02.92	08.50	26.78	37.43	14.89	03.61

*Stations 1, 4, 5, 6, 12, and 13 are sandy without any vegetation of seaweed or seagrass, **Station 2 is rocky substrate

Table 3: Biometric parameters of seaweed *Caulerpa prolifera* and seagrass *Halohpila stipulacea* at different sampling sites of Suez Canal site

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Caulerpa prolifera														
Percentage cover (%)	-	-	60±7	-	-	-	96±2	26±5	97±2	30±5	100±0	-	-	22±6
Total biomass g d wt. m^{-2}	-	-	541±20	-	-	-	1074±50	112±20	717±30	168±25	3008±100	-	-	391±25
Ash dry weight g d wt. m^{-2}	-	-	357±20	-	-	-	469±30	63±20	474±50	91±15	2092±90	-	-	304±35
Ash free dry weight g d wt. m^{-2}	-	-	184±25	-	-	-	605±30	49±10	243 ±15	77±10	916±35	-	-	87±20
Halophila stipulacea														
Percentage cover	-	-	-	-	-	-	-	-	-	23±5	-	-	-	76±6
Shoot density No.m^{-2}	-	-	-	-	-	-	-	-	-	72±17	-	-	-	163±20
Total biomass g d wt. m^{-2}	-	-	-	-	-	-	-	-	-	43±5	-	-	-	86±12
Ash dry weight g d wt. m^{-2}	-	-	-	-	-	-	-	-	-	12±3	-	-	-	22±5
Ash free dry weight g d wt. m^{-2}	-	-	-	-	-	-	-	-	-	31±5	-	-	-	64±8

Vegetation density and biomass: *C. prolifera* has a wide distribution at sites 3, 7, 8, 9, 10, 11 and 14 along the Suez Canal. While, it was absent at sites 1, 2, 4, 5, 6, 12 and 13. The percentage cover (%m^{-2}) ranged from 22% at site 14 to 100% at site 11 (Table 3) with lowest biomas 87 gm^{-2} (AFDW) and highest biomass of 916 gm^{-2} (AFDW) at

Table 4: Summary of Braun-Blanquet data on frequency of occurrence, abundance and density for seaweed *Caulerpa prolifera* and seagrass *Halophila stipulacea* along Suez Canal

	Caulerpa prolifera			*Halophila stipulacea*		
Status/site	Frequency	Abundance	Density	Frequency	Abundance	Density
1	-	-	-	-	-	-
2	-	-	-	-	-	-
3	0.80	3.80	3.20	-	-	-
4	-	-	-	-	-	-
5	-	-	-	-	-	-
6	-	-	-	-	-	-
7	1.00	5.00	5.00	-	-	-
8	0.60	2.40	1.50	-	-	-
9	1.00	5.00	5.00	-	-	-
10	0.40	1.80	1.60	0.35	1.80	1.30
11	1.00	5.00	5.00	-	-	-
12	-	-	-	-	-	-
13	-	-	-	-	-	-
14	0.40	1.40	1.30	0.90	4.40	4.10

both sites, respectively. At sites 8, 10 and 14 which showed less percentage cover and biomass of the seaweed where it was newly growing.

Seagrass *H. stipulacea* was recorded only at sites 10 and 14 in much localized patches with a percentage cover of 23 and 76% in their patches at both sites. The shoot density was higher at site 14, represented by 163±20 shoots m^{-2}; than site 10 which represented by 72±17 shoots m^{-2}. The total biomass of *H. stipulacea* at site 14 was 63.43 g m^{-2} (AFDW), while at site 10 was 30.14 g m^{-2} (AFDW).

The difference between percentage cover and biomass for the seaweed *C. prolifera* at different sites was statistically significant ($p<0.05$). Also, there was obvious difference between percentage cover, shoot density and biomass of the seagrass *H. stipulacea* at site 10 and site 14 ($p<0.05$). The difference may be attributed to the suitable environmental conditions at sites 8, 10 and 14 of low salinity and input of fresh water leading to low biometric data for *C. prolifera* and *H. stipulacea* and *C. racemosa* at these sites.

Braun-blanquet cover-abundance: Braun-Blanquet Cover-abundance (Table 4) showed that the seaweed *C. prolifera* occurred with the highest frequency, abundance and density both at the sites 7, 9 and 11; while, at other sites 8, 10 and 14, the seaweed was with lowest frequency, abundance and density.

Seagrass *H. stipulacea* was relatively frequent, dense and abundant at the site 14, than the site 10 (Table 4). The complete absence of seagrass from Lake Timsah and other sites is possibly due to the presence of unstable sediments covering the coast, which is unsuitable for the development of these plants, also, the anaerobic conditions at these sites and the discharge of herbicides and pesticides through Western Lagoon of Lake Timsah or other discharge points at Bitter Lakes.

Other ecological aspects and macroinvertebrates: The seaweed community at the sites 8, 10 and 14 was growing newly and consisted of delicate new blades. Recently grown rhizome apical meristems and central branches were also developing.

The blades of the investigated seaweed species were very clean, nearly free from macro-epiphytes. The microscopic examination showed very clean leaves in almost cases.

Invertebrate fauna of the seaweed community was less diverse than seagrass and sandy sites (Table 5). Thirty four species were recorded at different sampling sites. Only 14 species were recorded at *Caulerpa* sites, the most common species were mollusks *C. ruppelli*, *Trochus erythreus*, crustaceans and represented by the caridean shrimp *Alpheus audouini*, the crab *Eucrate crenata*, *Trapezia* sp. the amphipod *Gammarus* sp. and the isopod *Sphaeroma walkari*. Other habitats, seagrass and sandy areas were represented by 23 invertebrate species each. The most common species were the molluscs *Brachidontis variabilis*, *Cerithium erythraeonense*, *Modiolus auriculatus* and *M. tribulus*, crustaceans *Alpheus audouini*, *Gammarus* sp. *L. signata* and *Sphaeroma walkari* and polychaetes *Nereis persica*, *N. willeyi* and echinoderm *Ophicoma scolopendrina* at seagrass at sites 10 and 14. While at sandy areas, the most common species were mollusks *Bulla ampulla*, *Gafrarium pectinatum Murex tribulus*, *Venerupis pullastra* and *Tapes decussata*, crustaceans *Metapenaeus stebbingi* and the crab *Portunus pelagicus*, polychaetes *Branchiosyllis uncinigera*, *Glycinde bonhourei*, *Nereis persica*, *Phllodoce* sp. and *Syllis exilis* and echinoderms *Astropecten polyacanthus*, *Ophiactis savigyni* and *Ophicoma scolopendrina*.

Table 5: A list of invertebrate species of seaweed *Caulerpa prolifera*, seagrass community and sandy subtidal sites of Suez Canal during the present study

Species	*Caulerpa prolifera*	Seagrass	Sandy areas
Mollusca			
Brachidontes variabilis	-	++	+
Bulla ampulla	-	-	++
Cerithium erythraeonense	-	++	-
Cerithium ruppelli	++	+	-
Gafrarium pectinatum	-	+	++
Modiolus auriculatus	-	++	-
Murex tribulus	-	++	++
Pirenella cailliaudi	-	-	+
Venerupis pullastra	+	-	++
Tapes decussata	-	-	++
Trochus erythreus	++	+	+
Total	3	7	8
Crustacea			
Alpheus audouini	++	++	-
Atergatus roseus	-	+	-
Eucrate crenata	++	+	-
Gammarus sp.	++	++	-
Heteropanope laevis	+	-	-
Leucosia signata	+	++	-
Metapenaeus stebbingi	-	+	++
Portunus pelagicus	-	+	++
Sphaeroma walkari	++	++	-
Trapezia sp.	++	+	-
Total	7	9	2
Polychaeta			
Branchiosyllis uncinigera	-	-	++
Glycinde bonhourei	-	-	++
Glycinde multidens	++	-	+
Nereis persica	-	++	++
Nereis willeyi	-	+	++
Perinereis nuntia	-	+	-
Perinereis sp.	++	+	+
Phyllodoce sp.	++	+	++
Syllis exilis	-	-	++
Total	3	5	8
Echinodermata			
Astropecten polyacanthus	-	-	++
Hippocampus sp.	-	+	-
Ophiactis savignyi	-	-	++
Ophiocoma scolopendrina	+	++	++
Total	1	2	3
Total number of species	14	23	21

(-) Absent; (+) Rare, <1 up to 3 individuals 10 m^{-2}; (++) Common = 4 individuals 10 m^{-2};

DISCUSSION

Seagrass *Halophila stipulacea* known to occur in shallow waters along the western coasts of the Suez Canal (Fox, 1926; Lipkin, 1972), sometimes, this seagrass is intermingled to some extent with seagrass *H. uninervis* and seaweed *Caulerpa racemosa* (Lipkin, 1972). According to the present study, based on biometric parameters, the frequency of occurrence, abundance and density analyses, *H. stipulacea* is very rare seagrass now along the western coast of Suez Canal, which was recorded at two sites only from 14 sites investigated, while the other species *H. uninervis* was not recorded at all.

Now, *H. stipulacea* is replaced by the most frequent, dense competitive seaweed *C. prolifera*, which recorded at 50% of investigated sites, forming expanding dense meadows with percentage cover nearly 100% at many sites, especially at Bitter lakes. Last recent years after 2000, this species invaded and competed with the seagrass of Suez Canal, supplant them. This mainly happened; due to the competitive success of *C. prolifera* which seems to be related to some specific characters to this seaweed which are its big size, the high density of its populations (Meinesz and Hesse, 1991), its rapid growth (Komatsu *et al*., 1994), its high efficiency in dim light conditions (Gacia *et al*., 1994), its tolerance to the lack of severe nutrient limitation (Delgado *et al*., 1994), its wide temperature tolerance (Gacia *et al*., 1994) and its production of toxic secondary metabolites (Guerriero *et al*., 1992, 1993). The presence of these toxic secondary metabolites explains why *C. prolifera* is avoided by many of macro invertebrates. Many authors (Lemee *et al*., 1993; Dini *et al*., 1994, Fancour *et al*., 1994; Harmielin-Vivien *et al*., 1994; Pesandro *et al*., 1994) considered the discontinuity in the trophic chain from microbes to metazoans and ichtyofauna must exist in ecosystems dominated by *C. prolifera*.

The structure and flux of organic matter on the new ecosystem dominated by *C. prolifera* could be probably different to other Suez Canal infralittoral ecosystems. Its plant biomass is not very high conversely to its appearance and percentage cover. Productivity of *C. prolifera* is probably low. Studies (Romero *et al*., 1992; Pergent *et al*., 1994) confirm that *Caulerpa* being siphonalean algae, part of the new blades and stolon growth may be sustained by organic matter translocation from the senescent to the young, actively growing parts and it is not attributable to new growth. *C. prolifera* meadows do not act as a carbon sink, conversely to seagrass beds as confirmed by studies on *Posidonia oceanica* beds in Mediterranean (Romero *et al*., 1992; Pergent *et al*., 1994) according to a hypothesis would be that a great part of the produced organic matter is transferred to the water column as dissolved organic carbon. Thus, benthic production of *Caulerpa* beds would be exported to the pelagic system, conversely to what happen in seagrass meadows, where the transfer is from the pelagic to the benthos.

It was very clear during field investigation that macro-invertebrates community of *Caulerpa* was obviously low in biodiversity than seagrass and sandy habitat. This is due to its toxicity, which prevents grazing for some species (Guerriero *et al*., 1992, 1993) or to be a habitat for other biota (Lemee *et al*., 1993; Dini *et al*., 1994, Ferrer *et al*., 1994; Gianotti *et al*., 1994; Merino *et al*., 1994; Pesandro, *et al*., 1994; Talpin *et al*., 2005).

Sediments of the investigated sites were sandy ranging from very fine sand at *Caulerpa* sites to fine at sandy sites. Also, water was clearer at *Caulerpa* beds. *C. prolifera* have a great sediment retention capacity, favoring stabilization and organic enrichment of the environment (Walker and McComb, 1992). Due to this, plant density may affect the granulometric composition, shifting it to very fine sand and clay and increasing percentage of organic matter of the sediment, through slow down of water movements by seaweed blades.

The manifestation of a negative interaction of *C. prolifera* on *Halophila stipulacea* is consistent with other studies done using other species of seagrass (Talpin *et al*., 2005) and/or other members of the *Caulerpa* genus. The bulk of this prior work comes from the Mediterranean where exotic species of *Caulerpa* have been invading native seagrass beds (e.g., de Villele and Verlaque, 1995; Meinesz *et al*., 1993b; Boudouresque and Verlaque, 2002; Ceccherelli *et al*., 2002; Belsher *et al*., 2003). Also, Talpin *et al.* (2005). *C. prolifera* is an invasive species in Suez Canal, its interaction is similar to those observed in Mediterranean. In both Suez Canal and Mediterranean, interactions with *Caulerpa* sp. Resulted in a decrease in seagrass abundance under a variety of situations. In addition, anthropogenic alterations to coastal systems of Suez Canal may alter the outcome of these interactions.

In many conventions, signed by many countries on the environment and biological diversity. The issues regarding the exotic invasive organisms are taken into consideration. All countries ratifying the international conventions take responsibilities for controlling and monitoring the expansion of these species. Briefly speaking, Our goals in a combat against this invasive species is to monitor the spreading of the plant; forecasting the probable places that the species can colonize and to control already established colonies and to slow down development and spreading of the plant. Consequently, a current project in Suez Canal should be started by Ministry of Environment in order to protect biological diversity in our seas and to determine the present status of *C. prolifera* species along coasts as well as to plan action to undertake.

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