National Management Plan for the Genus *Caulerpa*

Photo by R. Woodfield, Merkel and Associates

Submitted to the Aquatic Nuisance Species Task Force

Prepared by the *Caulerpa* Working Group

October, 2005

Executive Summary

A variety of surveys have confirmed at least twenty-one species and varieties of *Caulerpa* with populations in different regions of the United States (U.S.). Three species of *Caulerpa* are thought to be invasive due to their historic and ongoing invasions of U.S. and foreign waters; *Caulerpa taxifolia* (Aquarium or Mediterranean strain), *Caulerpa brachypus* and *Caulerpa racemosa*.

In December 1999, the Aquatic Nuisance Species Task Force (ANSTF) established the *Caulerpa taxifolia* (Mediterranean strain) Prevention Committee. The Committee drafted the "Prevention Program for the Mediterranean strain of *Caulerpa taxifolia*" (Appendix A) which was an extension of the prior "Action plan to prevent the introduction and dispersal of the Mediterranean strain of *Caulerpa taxifolia* in U.S. waters".

In June 2000 divers detected *C. taxifolia* (Mediterranean strain) in Agua Hedionda Lagoon located in Carlsbad, CA and a second population in Huntington Harbor, CA. Divers first discovered non-native *C. brachypus* off the coast of southern Florida in 1999. Concerns have also been raised by scientists about *C. racemosa*, which has spread rapidly in the Mediterranean, but has not yet produced any problematic populations in U.S. waters.

Documented impacts of invasive *Caulerpa* species include competition with marine plants and macroalgae, direct and indirect impacts on marine invertebrates, direct and indirect impacts on marine vertebrates and economic impacts due to control costs and costs associated with ecosystem alteration. To date, eradication efforts for *C. taxifolia* in California have cost over $3.7 million in direct control costs, and over $500,000 have been allocated to study the effects of *C. brachypus* in Florida.

In 2002 the ANSTF recommended that a *Caulerpa* Working Group (CWG) be formed to provide input on the development of a National Management Plan (NMP) that addresses the genus *Caulerpa*. This NMP was developed with the input of the CWG and other *Caulerpa* experts to guide the ANSTF and other interested parties in managing *Caulerpa* species in U.S. waters.

A petition to list *Caulerpa taxifolia* (whole species) and *Caulerpa* (whole genus) was submitted to the U.S. Department of Agriculture Animal and Plant Health Inspection Service (USDA APHIS) on July 6, 2003. Currently the USDA APHIS is working on a pest risk assessment for the genus *Caulerpa* and will consider listing as Federal noxious weeds species of *Caulerpa* that are determined to pose a risk to the United States and that meet the International Plant Protection Convention definition of a quarantine pest.

The goals of this National Management Plan for the genus *Caulerpa* are:

1) Prevent the introduction and spread of *Caulerpa* species to areas in U.S. waters where they are not native.
2) Early detect, rapidly respond to and monitor *Caulerpa* species in U.S. waters where they are not native.
3) Eradicate *Caulerpa* populations, in waters to which they are not native, where feasible.
4) Provide long-term adaptive management and mitigate impacts of populations of *Caulerpa* species in U.S. waters where they are not native and where eradication is not feasible.
5) Educate and inform the public, agencies and policymakers to advocate for preventing the introduction and spread of *Caulerpa* species.
6) Identify research needs and facilitate research to fill information gaps.
7) Review, assess progress and revise the management plan and continue developing information to meet national management plan goals.

Table of Contents

Figures

Tables

Appendices

1. Purpose of this National Management Plan

The purpose of this National Management Plan (NMP) is to guide the Aquatic Nuisance Species Task Force (ANSTF) and other interested parties in managing invasive *Caulerpa* species already present in U.S. waters as well as species of the genus *Caulerpa* that may be introduced to U.S. waters to which they are not native. The ANSTF is an intergovernmental entity established under the Nonindigenous Aquatic Nuisance Prevention and Control Act of 1990 (Act, 6 U.S.C. 4701-4741), as amended by the National Invasive Species Act of 1996 (NISA). The ANSTF is co-chaired by the U.S. Fish and Wildlife Service (FWS) and the National Oceanic and Atmospheric Administration (NOAA). The ANSTF is responsible for coordination of national efforts to prevent the introduction and spread of aquatic invasive species. Among these responsibilities is to develop management plans for specific high-risk invasive species.

In December 1999, the ANSTF established the *Caulerpa taxifolia* (Mediterranean strain) Prevention Committee. The Committee drafted the "Prevention Program for the Mediterranean strain of *Caulerpa taxifolia*" (Appendix A) which was an extension of the prior "Action plan to prevent the introduction and dispersal of the Mediterranean strain of *Caulerpa taxifolia* in U.S. waters". Before the prevention plan could be implemented an invasive species of *Caulerpa* was discovered in two harbors of California's coastal region.

In June 2000, divers in a southern California lagoon found the first U.S. infestation of non-native *C. taxifolia* (Mediterranean strain). As a result of this discovery in southern California and the difficulty in distinguishing this strain from other *Caulerpa spp*., the ANSTF requested that the existing draft "Prevention Program for the Mediterranean strain of *Caulerpa taxifolia*" be further modified and expanded to a "National Management Plan (NMP)" for *Caulerpa* species.

In 2002 the ANSTF recommended that a *Caulerpa* Working Group (CWG) be formed to provide input on the development of a National Management Plan (NMP) that addresses the genus *Caulerpa* (ANSTF 2002). The intent of the NMP, which is more comprehensive than a prevention program, is to prioritize a variety of control strategies that federal, state, and local agencies, and the private sector can use, to prevent additional introductions, limit the spread of *Caulerpa spp*., eradicate *Caulerpa spp*., where feasible, and reduce the impacts of the existing populations. Control strategies may range from total eradication of entire populations to localized containment and prevention of further spread, to education programs targeting groups affected by or potentially involved in the algae's spread, to development of mitigation measures. In addition, the NMP identifies research needs to fill any gaps that may exist in the collective knowledge and ability to control and prevent the spread of *Caulerpa spp*. This plan incorporated the draft prevention plan submitted to the ANSTF for review as the basis for the development of this comprehensive NMP.

In February 2004, the CWG met to discuss the goals, objectives and priority actions of the NMP to manage the genus *Caulerpa* in U.S. waters. Subsequent to that meeting, CWG members ranked action items on their relevance to achieving the goals and objectives of the NMP in order to develop the primary, secondary and tertiary priorities for plan implementation (see Section 4). Other experts on aspects of the Genus *Caulerpa* also contributed to the development of this NMP. Appendix B lists all members of the *Caulerpa* Working Group as well as other contributors to the NMP, in 2004. The availability of the draft National Management Plan for the Genus *Caulerpa* was published in the Federal Register on August 17, 2005 (FR vol. 70, No. 158). All public comments were carefully considered for final version of this NMP. This, final version, of the NMP was unanomously approved by the ANSTF on October 20, 2005.

The goals of this NMP are as follows:

Goal 1: Prevent the introduction and spread of *Caulerpa* species to areas in U.S. waters where they are not native.

Goal 2: Early detect, rapidly respond to and monitor *Caulerpa* species in U.S. waters where they are not native.

Goal 3: To eradicate *Caulerpa* populations, in waters to which they are not native, where feasible.

Goal 4: Provide long-term adaptive management and mitigate impacts of populations of *Caulerpa* species in U.S. waters where they are not native and where eradication is not feasible.

Goal 5: Educate and inform the public, agencies and policymakers to advocate for preventing the introduction and spread of *Caulerpa* species.

Goal 6: Identify research needs and facilitate research to fill information gaps.

Goal 7: Review, assess progress and revise the management plan and continue developing information to meet national management plan goals.

2. Introduction to the Genus *Caulerpa*

Caulerpa is one of the most distinctive algal genera, identifiable solely on the basis of its habit (growth form) and internal morphology (Silva 2002). All species and subspecies of *Caulerpa* live in marine environments, but some can thrive in brackish lagoons (Silva 2004). Reports vary on the number of *Caulerpa* species from 70 (Meinesz 2002a) to approximately 100 (Dumay et al. 2002b), most of which inhabit warm waters.

The habit is typified by a creeping horizontal stem called a rhizome that produces tufts of colorless rhizoids (root-like structures) downward and photosynthetic branches or fronds (assimilators) upward (Silva 2002). The creeping rhizomes typically exhibit indeterminate growth (Collado-Vides and Robledo 1999). Photosynthetic assimilators assume many different forms, often with rows or whorls of leaf-like pinnules.

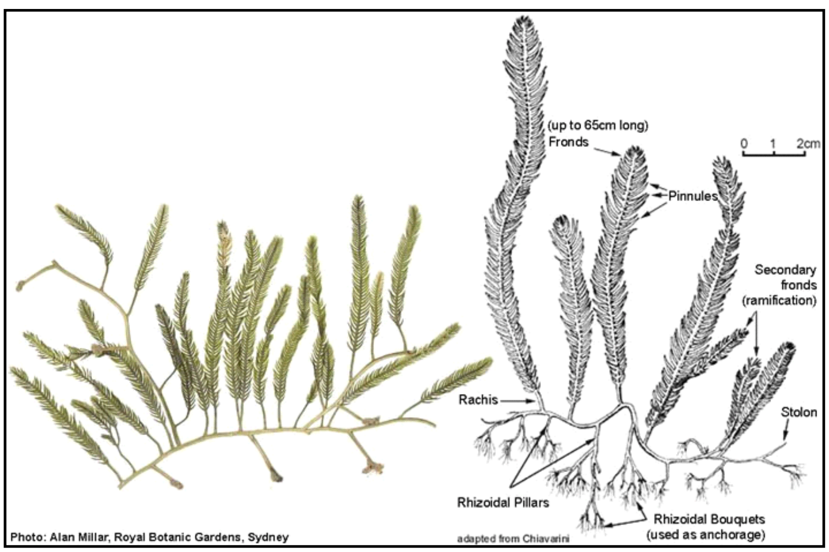
The variable morphology of certain species of *Caulerpa* is well known and experimental analysis has yet to definitively determine the relative influence of genetics versus environment in observed variation. On the other hand, many *Caulerpa* species show relatively little morphological variation.

In addition to its habit, *Caulerpa* has a distinctive suite of anatomical, cellular, and biochemical characters including reinforcement of the siphonous structure by cross-strands of fibrous algal wall material.

The algal body consists of a single cell siphonous structure that is not divided by cross-walls. A thin layer of cytoplasm, containing countless numbers of nuclei, chloroplasts and mitochondria, is closely pressed to the inside of the algal wall (Silva 2002). Asexual reproduction occurs readily by fragmentation of *Caulerpa* clones. Sexual reproduction is uncommon, but when it does occur almost all of the protoplast of a clone is converted into gametes (sex cells), effectively killing the clone.

Caulerpa clones divide labor, among organelles, into sugar production (photosynthetic) and energy storage (starch) functions (Silva 2002). Sugar production is facilitated in the assimilators via two photosynthetically active pigments that are unique to some green algae, siphonaxanthin and siphonein, which transfer energy directly to Chlorophyll A (Fujita, et al. 2003).

Figure 1. *Caulerpa taxifolia* exhibits a habit typical of the genus: a creeping rhizome produces rhizoids downward and photosynthetic branches (in this case bearing leaf-like pinnules) upward.



Members of the family Caulerpaceae can be particularly invasive (Davis et al. 1997). Distinct species of *Caulerpa* exhibit morphological and physiological forms that are adaptive to different environments. *Caulerpa* species that are endemic to lagoon habitats tend to have taller assimilators and longer rhizome distances between assimilators while those that thrive in high wave energy reef areas tend to express a more compact growth form (Collado-Vides and Robledo 1999). *C. racemosa* and *C. cupressoides*, which live in high light intensity environments, also tend to have lower chlorophyll content than species (i.e. *C. verticillata*) adapted to live in dense growths of other algae or sea-weed (Collado-Vides and Robledo 1999). While some *Caulerpa* species are primarily adapted to particular environments, others are generalist species that can grow in a wide variety of habitat types.

Beneficial Uses of *Caulerpa*

Certain species of *Caulerpa* have been used for centuries as food by the Japanese, Polynesians, Filipinos and Indonesians (Silva 2003). More recently live *Caulerpa* specimens have been distributed for ornamentation in marine aquariums and inadvertently transported with "live rock," which can harbor cryptic *Caulerpa* fragments (Frisch 2003). Pharmaceutical researchers have also expressed interest in the potential use of caulerpenyne in treatment of tumor cells (Barbier et al. 2001).

Invasive Characteristics of *Caulerpa*

Caulerpa, like other green algae such as *Bryopsis* and *Codium*, produces weedy strains, whose relationship to other strains is poorly understood (Silva 2002). Collado-Vides and Ruesink (2002) identify several biological traits that contribute to the invasive success of *C. taxifolia*. These traits, which may also apply to other *Caulerpa* species include:

- a high growth rate
- clonal growth form
- nutrient uptake from sediments
- tolerance of low water temperature
- lack of consumers

A high growth rate

A high growth rate allows some invasive *Caulerpa* species to rapidly colonize new areas thwarting attempts to economically control such species unless eradication is rapidly achieved. An examination of *C. taxifolia* patches growing in California's Huntington Harbor had a mean ± standard error number of stolon meristems (horizontal shoots or runners) of 555 ± 182 per square meter (Williams and Grosholz 2002). This high meristem density highlights the potential for proliferation across sediments and over other organisms (Williams and Grosholz 2002).

Clonal growth form

The ecological implications of clonal reproductive ability is that disturbances such as storms, or grazing by herbivores can result in *Caulerpa* fragments which are able to sprout and become new adults, from even tiny fragments (Smith and Walters 1999). Species capable of cloning may have a competitive advantage over many multicellular marine creatures that reproduce sexually (Vroom and Smith 2001). Successful spread via fragmentation varies among *Caulerpa* species and appears to be a critical factor in the ability to rapidly colonize new areas (Smith and Walters 1999).

Nutrient uptake from sediments

Unlike most macroalgae, which anchor to sediments and derive nutrients from the water column (Collado-Vides and Ruesink 2002) species of the genus *Caulerpa* have rhizoids that embed into sediments and take nutrients from the sediments. For instance, the rhizoids of *C. taxifolia* mimic the roots of vascular plants and have been observed to directly take carbon, nitrogen and phosphorus from substrata (Chisholm et al. 1996). The superior access to nutrients provided by rhizoids may make *Caulerpa* species superior competitors in nutrient limited environments (Williams 1984a). Not only are the rhizoids of *C. taxifolia* and other species of the genus capable of deriving nutrients directly from sediments, but their rhizoids also form associations with a bacterium that may be capable of fixing gaseous nitrogen (Collado-Vides and Ruesink 2002).

<u>Tolerance of low water temperature</u>
Caulerpa species are among the most widespread algae occurring in tropical and subtropical waters, as well as in certain temperate waters (Silva 2003). The ability of some *Caulerpa* species to survive relatively low water temperatures may allow those species to exploit new niches if they are introduced. A study conducted in 2001 showed that twelve out of the fourteen species of *Caulerpa* species that were available for sale in southern California have native distributions that extend into temperate waters (Frish 2003).

<u>Lack of consumers</u>
Tropical fish such as parrotfish (Scaridae) and surgeonfish (Acanthuridae) have adapted to successfully graze on *Caulerpa* species (Paul and Hay 1986) as well as some invertebrate herbivores (Walters per. comm. 2004). Conversely, common marine invertebrates and vertebrates in temperate regions have been found to be highly susceptible to a toxic compound found in *Caulerpa* (caulerpenyne) and will not graze on *Caulerpa* (Paul and Fenical 1986).

2.1. Species of *Caulerpa* which are native to U.S. waters

Many *Caulerpa* species are native to warm coastal waters of the North, Central and South America. On the Pacific coast of North America, no indigenous species of *Caulerpa* are found north of Guadalupe Island, at 29º N (Silva 2003) and only one species *Caulerpa vanbosseae* is found this far north (Silva per. comm. 2004). On the Atlantic coast of North America some *Caulerpa* species reside as far north as Onslow Bay, North Carolina, at latitude 34ºN (Silva 2003).

Figure 2. On the Pacific coast of North America indigenous populations of *Caulerpa* only exist as far north as Guadalupe Island.

Both Florida and Hawaii are known to have many resident species of *Caulerpa* in their coastal waters. Surveys have confirmed at least twenty-one species and varieties of *Caulerpa* with populations in different regions of the U.S., as displayed in Table 1 (Frisch 2003, LaPointe per. comm. 2004 and Walters per. comm. 2004).

Research on resident *Caulerpa* species in U.S. waters is relatively recent and the phylogenetic relationship between species is not well documented. In addition, the vast coastal regions of U.S. states and territories encompass a wide variety of marine ecosystems. For instance, a species of *Caulerpa* that is native to the southeast coast of Florida may be a natural part of the ecosystem and may not show invasive characteristics in its native range. However a *Caulerpa* species that is native in the southeast may have no predators on the Pacific coast and may be invasive if introduced. Due to the scientific uncertainty about the origins of resident *Caulerpa* species this NMP will avoid characterizing species as native or non-native and will attempt to focus on potential for invasiveness in waters of the U.S.

Table 1. Many *Caulerpa* species are found in coastal waters of U.S. states and territories.

Species	***Reference***	***Location found***
C. ambigua	Peyton 2004	Guam, Saipan, Hawaii
C. antoensis	Peyton 2004	Guam, Saipan, Hawaii
C. ashmedii	Walters 2004	Florida Keys, Tampa Bay region
C. brachypus var. parvifolia	(a) Lapointe 2004 (b) Peyton 2004	(a) East coast of central FL (b) Guam
C. cupressoides	(a) Walters 2004, Paul 2002, Collado-Vides and Robledo 1999 (b) Peyton 2004	(a) Florida Keys (b) Guam, Saipan, Hawaii
C. elongata	Peyton 2004	Saipan, Hawaii
C. fastigiata	Lapointe 2004	Coastal waters of FL
C. filicoides	Peyton 2004	Guam, Saipan
C. floridiana	Lapointe 2004	Coastal waters of FL
C. freycinetii	Peyton 2004	Guam
C. languinosa	Walters 2004, Paul 2002, Paul and Hay 1986	Florida Keys
C. lentillifera	Peyton 2004	Guam, Saipan, Hawaii
C. macrophysa	(a) Walters 2004 (b) Peyton 2004	(a) Florida Keys (b) Hawaii
C. mexicana	(a) Walters 2004, Paul 2002 (b) Peyton 2004	(a) Florida Keys, Southwest coast of FL, Tampa Bay region (b) Hawaii
C. microphysa	Frisch 2003	Waikiki Beach HI, Florida Keys, Miami Beach FL
C. nummularia	Peyton 2004	Hawaii
C. ollivieri	Lapointe 2004	Coastal waters of FL
C. paspaloides	Walters 2004	Florida Keys, Tampa Bay region
C. peltata	(a) Lapointe 2004 (b) Frisch 2003 (c) Peyton 2004	(a) Coastal waters of FL, (b) Waikiki Beach HI (c) Guam, Saipan
C. pinnata	Peyton 2004	Hawaii
C. prolifera	(a) Walters 2004 (b) Frisch 2003	(a) East coast of central FL, Florida Keys, Southwest coast of FL, Tampa Bay region (b) Onslow Bay NC
C. racemosa	(a) Walters 2004, Paul and Hay 1986 (b) Frisch 2003 (c) Peyton 2004, UHM 2004	(a) East coast of central FL, Florida Keys, Southwest coast of FL, Tampa Bay region (b) Oahu HI, Hawaii HI, Maui HI (c) Guam, Saipan, Hawaii
C. racemosa v. macrophysa	Frisch 2003	Oahu, HI
C. racemosa v. peltata	Frish 2003	Oahu, HI
C. serrulata	(a) Frisch 2003, UHM 2004 (b) Peyton 2004	(a) Kauai HI, Oahu, HI (b) Guam, Saipan, Hawaii
C. sertularioides	(a) Walters 2004 (b) Frisch 2003 (c) Peyton 2004	(a) East coast of central FL, Florida Keys, Southwest coast of FL, Tampa Bay region (b) Halona/Kaloko HI, Hanauma Bay HI, Maui HI, Oahu HI, Hawaii HI, Pearl Harbor HI, Miami FL (c) Guam, Saipan, Hawaii
C. taxifolia	(a) Frisch 2003, UHM 2004 (b) Peyton 2004	(a) Hawaii HI, Kauai HI, Oahu HI (b) Guam, Saipan, Hawaii
C. urvilleana	Peyton 2004	Guam, Saipan
C. verticilata	(a) Walters 2004 (b) Peyton 2004, UHM 2004	(a) Florida Keys (b) Guam, Hawaii
C. webbiana	(a) Lapointe 2004 (b) Frisch 2003 (c) Peyton 2004	(a) Coastal waters of FL (b) Oahu HI, Honalulu HI (c) Guam, Hawaii

2.2. Species of *Caulerpa* that may be introduced to U.S. waters to which they are not native

Three *Caulerpa* species are of particular concern due to their historic and ongoing invasions of U.S. and foreign waters; *C. taxifolia*, *C. brachypus* and *C. racemosa*. The cosmopolitan introduction and spread of *C. taxifolia* has led to wide documentation of its history, biology, ecology and impacts on native ecosystems (see Appendix C). In Florida concern has recently been raised about the introduction of *C. brachypus* (FDEP 2003), which has produced large patches that overgrow sponges, hard corals, octocorals, and even other algal species off the coast of northern Palm Beach County (Lapointe and Barile 2001). Concerns have also been raised about *C. racemosa*, which in the Mediterranean, has spread to eleven countries since 1991 (Engrave 2003). The following sections focus on the current, available knowledge about the invasive characteristics of *Caulerpa* species of concern. Other *Caulerpa* species could also be invasive in U.S. waters, but they remain largely unstudied.

2.2.1. *Caulerpa taxifolia*

A detailed literature review of *C. taxifolia* is presented in Appendix C. The brief introduction to *C. taxifolia* that follows will refer to the species unless the Mediterranean strain is clearly indicated. *C. taxifolia* naturally occurs in tropical and subtropical waters of the Caribbean, Indonesia, Southeast Asia, Australia, and Hawaii (Guiry and Dhonncha 2004). In Australia, it naturally occurs in habitats ranging from sheltered bays to coasts and reef edges with high energy wave action (Phillips et al. 2002). In its native range *C. taxifolia* is limited by winter sea surface temperatures below 19°C (Meinesz 2002a). This species reproduces both asexually and sexually, primarily growing in small clumps.

Recognized forms and varieties (Guiry and Dhonncha 2004)

Caulerpa taxifolia f. *asplenoides* (Greville) Weber-van Bosse
Caulerpa taxifolia var. *crassifolia* C. Agardh
Caulerpa taxifolia f. *interrupta* Svedelius
Caulerpa taxifolia f. *tristichophylla* Svedelius

Figure 3. *Caulerpa taxifolia* showed vigorous growth, prior to treatment in California.

Regional distribution (Guiry and Dhonncha 2004)

Europe, Mediterranean Basin, Caribbean Basin, West Africa, East Asia, South-east Asia, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

Environmental tolerance

The Mediterranean strain of *C. taxifolia* is adapted to a wide range of environments. It can survive in habitats that range from 1 to 30 m in depth (Makowa 2000). The photosynthetic pigments siphonaxanthin and siphonein, which facilitate photosynthesis in *C. taxifolia*, have been found to be more evident in marine plants that live in deeper habitats (SZAD 2003). It is possible that the presence of these pigments allows *C. taxifolia* to thrive in lower light levels and at greater depth then other macroalgae. These special adaptations may also allow *C. taxifolia* to be a superior competitor with other macroalgae.

In a controlled lab environment, the upper lethal temperature was between 31.5-32.5°C (88.7-90.5ºF) and the lower was between 9°C-10°C (48.2-50ºF) [Komatsu et al. 1997]. In the same experiment the alga survived without growth between 10-12.5°C (50-54.5ºF). New stolons developed at 15°C (59ºF) and new fronds developed at 17.5°C (63.5ºF) [Komatsu et al. 1997]. Under laboratory conditions the Mediterranean strain of *C. taxifolia* has also been shown to survive for three months at 10°C, even at low light levels (Meinesz 1999). This laboratory work is supported by the observation that when introduced to the Sea of Japan in 1992, *C. taxifolia* failed to establish where the mean surface water temperature dropped to 9.8ºC for the coldest two months of the year (Komatsu et al. 2003).

While anecdotes exist that native strains of *C. taxifolia* are not particularly cold tolerant and the Mediterranean strain has unusual cold tolerance due to aquarium cultivation, few studies have been performed on the cold tolerance of native strains (Chisholm et al. 2000). One group of researchers that did test wild *C. taxifolia* samples from Moreton Bay, Australia found that it had a similar cold tolerance to that reported for the Mediterranean strain. Under laboratory conditions, samples of *C. taxifolia* from Moreton Bay were able to survive, and recover fully from 4-6 weeks of exposure to 11 ºC (52 ºF) [Chisholm et al. 2000].

Experiments have demonstrated that as clones become more dense growth of *C. taxifolia* clone density increases, primarily due to shelter effects from water movement (Piazzi and Ceccherelli 2002). This result suggests that the invasive characteristics of *C. taxifolia* may increase with colonization time (Piazzi and Ceccherelli 2002).

C. taxifolia can grow particularly well in areas with substrate containing enriched organic matter (Chisholm et al. 1996). The ability of *C. taxifolia* rhizoids to take up carbon, nitrogen and phosphorus from substrata may explain its ability to dominate in areas with oligotrophic water and eutrophic substrata (Chisholm et al. 1996). Furthermore, data indicate that growth of *C. taxifolia* enhances nutrient availability in high carbon/nitrogen ratio sediments by a process in which photosynthetic products (sugars) leaked from rhizoids alter sedimentary bacterial ecology leading to a chain reaction that results in increased nitrogen availability (Chisholm and Moulin 2003). A proportion of the nitrogen produced by this bacterial chain reaction can then be taken up by the rhizoids of *C. taxifolia*, enhancing its growth (Chisholm and Moulin 2003). This mutualistic association between bacteria and *C. taxifolia* can explain the ability of *C. taxifolia* to spread into areas with low nutrient water and sediments high in organic matter such as dead *Posidonia* mat areas in the Mediterranean (Chisholm and Moulin 2003). In addition, studies of the light requirements of *C. taxifolia* in the Mediterranean imply that *C. taxifolia* survival resulting from photosynthesis should be limited to a depth of 29 m in summer and 10 m in winter (Chisholm and Jaubert 1997). Another study has suggested a discrepancy between photoautotrophic supported growth and observed growth implying that *C. taxifolia* is able to continue growth by absorbing carbon from the substrate rather then through photosynthesis alone (Chisholm and Jaubert 1997).

Reproduction

Native, tropical *C. taxifolia* reproduce sexually and asexually. When sexual reproduction occurs, native *C. taxifolia* algae release male and female gametes. While sexual reproduction of *C. taxifolia* in tropical habitats is common, in the Mediterranean *C. taxifolia* has only been observed to release male gametes (Zuljevic and Antolic 2002b). A new *C. taxifolia* colony can develop from a small fragment of stolon, rhizoid, frond, or pinnule (Smith and Walters 1999, Zuljevic and

Antolic 2002a). In a laboratory setting a single frond has been shown to develop into a new plant in as little as ten days at 25°C (77 ºF) [Zuljevic and Antolic 2002a].

Defense mechanisms

Like most species of *Caulerpa*, *C. taxifolia* produces caulerpenyne (CYN), a toxic terpenoid, which is found in particularly high concentrations in the invasive Mediterranean clone (Paul 2002). *C. taxifolia* also contains a host of potentially toxic distasteful terpenoids and other nitrogenous compounds that may fulfill defensive purposes broadly within species of Caulerpales (Lemee et al. 1993).

When a *C. taxifolia* clone is wounded, as in by feeding by an herbivore, caulerpenyne is enzymatically transformed to oxytoxins, which are likely to be an even more potent chemical defense (Paul 2002). Many tropical herbivores are not deterred by *Caulerpa* chemical defenses, but non-adapted temperate species could be more affected by these compounds (Paul 2002).

Research in Australia has found that water temperatures in excess of 19ºC and depths of less then 5 meters are the optimum conditions for caulerpenyne production (Millar 2002), but many additional factors can also influence caulerpenyne production. In the Mediterranean, Amade and Lemee (1998) found that caulerpenyne content varies seasonally, with the lowest concentrations in winter and spring followed by a sharp increase between June and July. Furthermore caulerpenyne content was highest in specimens at a depth of 5 m and decreased from 5 m to 30 m deep with the lowest concentrations being found at the greatest depths (Amade and Lemee 1998).

Natural controls

In tropical regions marine vertebrates and invertebrates are known to feed on *C. taxifolia*, but few temperate species have been identified as consumers. In New South Wales, Australia researchers have confirmed through direct observation and sampling (Sakker per. comm.) that a fish, the luderick (*Girella triccuspidata*), feeds on *C. taxifolia*. Anecdotal reports of surgeon fish (*Acanthurus* spp.) eating *C. taxifolia* have also been received in New South Wales (Sakker per. comm.).

In the Mediterranean two species of ascoglossan molluscs, *Oxynoe olivacea* and *Lobiger serradifalci* have been found to feed on *C. taxifolia* (Thibaut and Meinesz 2000). Rather then effecting grazer control of *C. taxifolia* populations, ascoglossan herbivores create thallus fragments, which is a principle method of dispersal and population spread (Zuljevic et al. 2001). These two species of molluscs are also routinely found feeding on a different *Caulerpa* species, *Caulerpa prolifera*, in the Mediterranean (Thibaut and Meinesz 2000).

Figure 4. *Caulerpa brachypus* patches are growing on coral in FL.

2.2.2. *Caulerpa brachypus*

C. brachypus was first described from Kagoshima Prefecture in Japan, but it is known to occur naturally throughout the Indo-Pacific oceans (Millar 2004 per. comm.)

C. brachypus exhibits a typical *Caulerpa* habit with fronds emanating upward and rhizoids downward from a horizontal stolon. Individual fronds can reach up to a height of 5 cm (FDEP 2003). The photosynthetic assimilators typically form a tongue like, green ribbon sometimes with a yellowish colored edge (IPTEK 2004).

Recognized forms and varieties (Guiry and Dhonncha 2004)
Caulerpa brachypus var. *parvifolia* (Harvey) A.B. Cribb
Caulerpa brachypus var. *brasiliana* A.B.Joly & Semir
Caulerpa brachypus var. *nordestina* A.B.Joly & Semir
Caulerpa brachypus f. *parvifolia* (Harvey) Cribb
Caulerpa brachypus var. *mauritiana* (Børgesen) Børgesen
Caulerpa brachypus f. *exposita* Børgesen

Regional distribution (Guiry and Dhonncha 2004)
Caribbean Basin, East Asia, South-east Asia, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

U.S. distribution (LaPointe & Barile 2001)
Has recently been found off the east coast of Florida (LaPointe & Barile 2001).

Environmental tolerance
C. brachypus is known to be a warm-temperate to tropical species (Millar, personal communication, July 5, 2004). Diver surveys off the coast of southern Florida have collected *C. brachypus* at depths ranging between 25-47 m (82-154 ft.), at temperatures ranging from 24-28ºC (75-82ºF) and water visibility from 27-50 m (89-164 ft.) [Lapointe and Barile 2001]. The same survey found that *C. brachypus* was most abundant at a 42 m (138 ft.) deep site.

The high photosynthetic yield and low respiration rate of *C. brachypus* is indicative of a shade algae (Lapointe and Yentsch 2003). The efficient photosynthesis exhibited by *C. brachypus* allows colonies to continue to meet nutritional requirements despite dense crowding on reefs sites ranging from 60-90 percent cover of *C. brachypus* in the spring, summer and fall (Lapointe and Yentsch 2003).

Reproduction
C. brachypus is thought to have reproductive characteristics similar to other typical members of the genus *Caulerpa* (Millar per. comm. 2004).

Defense mechanisms
Preliminary evidence from studies on the coast of Florida suggests that caulerpenyne in *C. brachypus* may not be present in sufficient quantities to deter grazing by generalist herbivores such as parrotfish (Family: Scaridae) [Lapointe and Yentsch 2003]. None the less, the amount of caulerpenyne present may act as an important activated defense compound (Lapointe and Yentsch 2003).

2.2.3. *Caulerpa racemosa*

C. racemosa is reported to be widely distributed in tropical seas (Piazzi et al. 2001b). Taxonomic and ecological studies have demonstrated that there are three distinct *C. racemosa* varieties coexisting in the Mediterranean Sea (Piazzi et al. 2001a). *C. racemosa* has also been reported along the coasts of portions of the U.S. (Frisch 2003) as well as Israel, Syria and Turkey (TDN 1998).

Genetic analysis has demonstrated that unlike *C. taxifolia*, which most likely came from a single source, the *C. racemosa* populations in the Mediterranean Sea show genetic variation, associated with relaxed selection, typical of naturally occurring populations (Fama et al. 2000a).

Figure 5. *Caulerpa racemosa* can grow rapidly in some environments.

<u>Recognized forms and varieties (Guiry and Dhonncha 2004)</u>

Caulerpa racemosa (Forsskål) J. Agardh
Caulerpa racemosa f. *requienii* (Montagne) Weber van Bosse
Caulerpa racemosa var. *clavifera* (Turner) Weber-van Bosse
Caulerpa racemosa f. *microphysa* Weber-van Bosse
Caulerpa racemosa f. *reducta* Børgesen
Caulerpa racemosa var. *laetevirens* (Montagne) Weber-van Bosse
Caulerpa racemosa var. *lamourouxii* (Turner) Weber-van Bosse
Caulerpa racemosa var. *occidentalis* (J. Agardh) Børgesen
Caulerpa racemosa var. *peltata* (Lamouroux) Eubank
Caulerpa racemosa var. *turbinata* (J. Agardh) Eubank
Caulerpa racemosa var. *macrophysa* (Sonder ex Kützing) W.R. Taylor
Caulerpa racemosa f. *condensata* Weber-van Bosse
Caulerpa racemosa var. *uvifera* (C. Agardh) J. Agardh
Caulerpa racemosa var. *chemnitzia* (Esper) Weber-Van Bosse
Caulerpa racemosa var. *corynephora* (Montagne) Weber-van Bosse
Caulerpa racemosa var. *gracilis* (Zanardini) Weber-van Bosse
Caulerpa racemosa var. *macra* Weber-van Bosse
Caulerpa racemosa var. *macrodisca* (Decaisne) Weber-van Bosse
Caulerpa racemosa var. *imbricata* (Kjellman) Eubank
Caulerpa racemosa var. *microphysa* (Weber-van Bosse) Reinke
Caulerpa racemosa var. *nummularia* (Harvey ex J. Agardh) D. Dixit
Caulerpa racemosa var. *laetevirens* f. *cylindracea* (Sonder) Weber-van Bosse
Caulerpa racemosa f. *laxa* (Greville) Weber-van Bosse
Caulerpa racemosa f. *compressa* Weber-van Bosse
Caulerpa racemosa f. *semifalcata* Chauhan & Thivy
Caulerpa racemosa f. *simplicissima* Børgesen
Caulerpa racemosa f. *intermedia* Weber-van Bosse
Caulerpa racemosa f. *complanata* (J. Agardh) Weber-van Bosse
Caulerpa racemosa var. *intermediata* P. Anand
Caulerpa racemosa var. (*lamourouxii*) f. *requienii* (Montagne) Weber-van Bosse
Caulerpa racemosa var. (*clavifera*) f. *macrophysa* (Sonder ex Kützing) Weber-van Bosse
Caulerpa racemosa f. *occidentalis* (J.Agardh) Nizamuddin

Caulerpa racemosa f. *turbinata* (J. Agardh) Weber-van Bosse
Caulerpa racemosa var. *zeyheri* (Kützing) Weber-van Bosse
Caulerpa racemosa var. *exigua* (Weber-van Bosse) Eubank
Caulerpa racemosa var. *cylindracea* (Sonder) Verlaque, Huisman & Boudouresque
Caulerpa racemosa var. *disticha* V.J. Chapman

Regional distribution (Guiry and Dhonncha 2004)
Europe, Mediterranean Basin, Caribbean Basin, West Africa, Eastern and Western South America, Western, North and Central America (Islas Revilla Gigedo), South-east Asia, Pacific Islands, Archipelago, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

Environmental tolerance
C. racemosa has been collected at depths up to 47 meters (154 ft.), with water visibility to 27 meters (138 ft.) and a temperature of 19ºC (66ºF) off the coast of southern Florida (Lapointe and Barile 2001). Physiological adaptations have been noted in *C. racemosa*, when comparing shallow samples to those collected at a depth of 37 meters, which allow it to tolerate lower light levels at greater depths (Riechert and Dawes 1986).

Growth of *C. racemosa* has been shown to increase as density increases, but to a lesser degree than with *C. taxifolia* (Piazzi and Ceccherelli 2002). However, competition studies suggest that where *C. taxifolia* and *C. racemosa* occur together, *C. racemosa* will be the favored species (Piazzi and Ceccherelli 2002).

In laboratory conditions, *C. racemosa* stopped growing when salinity was reduced to 20 ppt, but did not die after twenty days (Carruthers et al. 1993). *C. racemosa* can withstand short-term exposure to salinity as low as 20ppt, and may be able to begin growth if salinity levels rise after that exposure (Carruthers et al. 1993).

C. racemosa has been observed to grow over rock substratum and dead mats of *Posidonia oceanica* (seagrass) covering both types of substrata in six months from the start of the invasion (Piazzi et al. 2001b). However, biomass production was higher on seagrass mats, suggesting that this is a favorable substratum (Piazzi et al. 2001b). Meadows of *P. oceanica* can also be invaded by *C. racemosa* (Piazzi et al. 2001b).

Reproduction
C. racemosa is known to reproduce both sexually and asexually by random fragmentation (Renoncourt and Meinesz 2002). Propagation through vegetative fragmentation is an important mode of asexual reproduction (Ceccherelli and Piazzi 2001). *C. racemosa* fragments show no decrease in ability to establish even after drifting for ten days (Ceccherelli and Piazzi 2001). In the Mediterranean asexual reproduction is considered to greatly contribute to the spread of *C. racemosa* (Ceccherelli and Piazzi 2001). *C. racemosa* has also been observed to produce vegetative propagules which may explain the disappearance of *C. racemosa* meadows during the winter and their reappearance by the end of spring (Renoncourt and Meinesz 2002).

Research findings suggest that *C. racemosa* will out-compete *C. taxifolia* and that the presence of *C. taxifolia* colonies can facilitate the spread of *C. racemosa* (Ceccherelli 2002). In the same environment *C. racemosa* has the ability to spread faster than *C. taxifolia* indicating a very high invasive potential for *C. racemosa* (Piazzi et al. 2001).

Defense mechanisms
Some specimens of *C. racemosa* have been found to contain the defensive compound caulerpenyne (Cimino and Ghiselin 1998). In Discovery Bay, Jamaica research suggests that *C. racemosa* is resistant to fish grazing by dominant reef fish such as parrot fish (Scaridae), but can not deter grazing by the long-spined urchin (*Diadema antillarum*) [Morrison 1988].

Natural controls
The long-spined urchin (*Diadema antillarum*), which is endemic to the Caribbean, readily consumes *C. racemosa* (Morrison 1988), although it is not a preferred food (Williams per. comm. 2004). A mass mortality of long-spined urchins, in the 1980s, led to an increase in the abundance of *C. racemosa* in Discovery Bay, Jamacia suggesting the importance of this herbivory pressure in regulating *C. racemosa* abundance (Morrison 1988). A sacoglossan mollusk (*Lobiger serradifalci*) has also been observed feeding on wild populations of *C. racemosa* off the coast of Venezuela (Cimino and Ghiselin 1998). This sea slug, *L. serradifalci*, modifies the chemical defenses of *C. racemosa* into the metabolites oxytocin-1 and oxytocin-2 for its own use as a deterrent to predation (Cimino and Ghiselin 1998).

2.2.4. Other potentially invasive species of Caulerpa

Caulerpa prolifera

C. prolifera has shown characteristics that might be linked to invasiveness. The growth rate of *C. prolifera* is comparable to that of multicellular higher plants (Chen and Jacobs 1966). In a laboratory setting [12-light/12-dark cycle, 24ºC (75ºF)] *C. prolifera* rhizomes elongated at a rate of 2 to 8 mm/day and produced a new cluster of rhizoids every day (Chen and Jacobs 1966). Fragments are most successfully established by rhizomes as fronds rarely survive fragmentation (Smith and Walters 1999).

Figure 6. *Caulerpa prolifera* has shown characteristics that might be linked to invasiveness.

Recognized forms (as listed in Guiry and Dhonncha 2004)
Caulerpa prolifera f. *obovata* J.Agardh
Caulerpa prolifera f. *zosterifolia* Børgesen

Regional distribution (as cited in Guiry and Dhonncha 2004)
Europe, Mediterranean Basin, Caribbean Basin, West Africa, Eastern South America, South-east Asia.

U.S. distribution
East coast of central Florida, Florida Keys, Southwest coast of Florida, Tampa Bay Region (Walters 2004) and Onslow Bay NC (Frisch 2003).

Environmental tolerance
C. prolifera has been observed growing in a coastal lagoon, in the south-east Iberian peninsula, with water salinity that ranged from 42 to 47 ppt (compared to 36-38 ppt in the Mediterranean) [Terrados and Ros 1991]. In the same lagoon, the lower temperature limit of *C. prolifera* was 10ºC in February and 20ºC in June; the high limit remained constant near 35ºC (Terrados 1991).

Fertilization experiments indicate that growth of *C. prolifera* is primarily limited by nitrogen (Terrados 1991). This finding is supported by observation that *C. prolifera* grows best in mud, although it will also grow on sand, clay or rock (Terrados and Ros 1991).

C. prolifera can grow at least as deep as 50 meters (Johnston 1969). Measurement of photosynthetic characteristics of *C. prolifera*, compared to other macroalgae suggest that *C. prolifera* is adapted to photosynthesize in low light environments (Terrados and Ros 1992). Growth and nitrogen concentration increase have been observed to increase in *C. prolifera* up to a light intensity of 2,500 lux[1], while further increases in light intensity caused a significant decrease in growth and nitrogen concentration (Khaleafa et al. 1982). Specimens that grow in deep water have greater internodal length possibly allowing them to prevent self-shading and further increasing their shade tolerance (Johnston 1969).

Reproduction
C. prolifera has the capacity to vegetatively form a new thallus (rhizome) from small fragments that drift in water, allowing it to rapidly colonize new areas (Terrados and Ros 1991).

Natural controls
A shelled sacoglossan mollusk (sea slug) *Ascobulla fragilis* feeds on *C. prolifera* by extracting plant sap and modifing the extracted caulerpenyne into an oxytocin compound for use as a secondary defense compound (Cimino and Ghiselin 1998). Two other slugs *Oxynoe olivacea* and *Lobiger serradifalci* also feed on *C. prolifera* and modify its defense compounds into oxytocins for their own defense (Cimino and Ghiselin 1998). In the Caribbean Basin, another sacoglossan mollusk, *Elysia subornata* lives and feeds on *C. prolifera* (Cimino and Ghiselin 1998).

Natural populations off the Canary Islands do not grow fast enough to overcome grazing at depths shallower than 15 meters, limiting their spread (Johnston 1969).

Caulerpa cupressoides
Caulerpa cupressoides has been shown to exhibit very fast rates of growth. Stolon elongation rates can be very high ranging from 0.22 to 7.51 cm/day (*C. cupressoides* v. *lycopodium* and *C. cupressoides* f. *elegans*) in relatively deep water (Williams et al. 1985). The ability of *C. cupressoides* to modify morphological and physiological performance allows it to succeed in varied environments (Collado-Vides and Robledo 1999). Of the seven species of *Caulerpa* studied in the Mexican Caribbean only *C. cupressoides* exhibited the variable morphology required to thrive in both high energy reef and lagoon habitats. Photosynthetic efficiency was also maximal in *C. cupressoides*, but varied with the *in situ* environment (Collado-Vides and Robledo 1999).

[1] Lux: a unit of illumination equal to the direct illumination on a surface that is everywhere one meter from a uniform point source of one candle intensity or equal to one lumen per square meter (Merriam-Webster 2004).

Figure 7. ***Caulerpa cupressoides*** **has been shown to exhibit very fast rates of growth.**

Recognized forms and varieties (as listed in Guiry and Dhonncha 2004)

Caulerpa cupressoides var. *lycopodium* Weber-van Bosse
Caulerpa cupressoides f. *amicorum* (Harvey) Weber-van Bosse
Caulerpa cupressoides f. *disticha* Weber-van Bosse
Caulerpa cupressoides f. *elegans* (P. Crouan & H. Crouan) Weber-van Bosse
Caulerpa cupressoides var. *mamillosa* (Montagne) Weber-van Bosse
Caulerpa cupressoides var. (*lycopodium*) f. alternifolia (P. Crouan H. Crouan) Weber-van Bosse
Caulerpa cupressoides var. *plumarioides* Børgesen
Caulerpa cupressoides var. *serrata* (Kützing) Weber-van Bosse
Caulerpa cupressoides var. *turneri* Weber-van Bosse
Caulerpa cupressoides var. *ericifolia* (Turner) Weber-van Bosse
Caulerpa cupressoides var. *flabellata* Børgesen
Caulerpa cupressoides f. *intermedia* Weber-van Bosse
Caulerpa cupressoides f. *nuda* Weber-van Bosse
Caulerpa cupressoides f. mamillosa
Caulerpa cupressoides f. *gardineri* Weber-van Bosse
Caulerpa cupressoides var. *disticha* Weber-van Bosse
Caulerpa cupressoides f. *denudata* Weber-van Bosse

Regional distribution (as cited in Guiry and Dhonncha 2004)

Europe, Caribbean Basin, West Africa, East Asia, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Amirante Islands, Australia and New Zealand.

U.S. distribution

Florida Keys (Walters 2004), Guam, Saipan and Hawaii (Peyton 2004).

Environmental tolerance

This species has demonstrated the ability to grow rapidly under tolerable conditions. *C. cupressoides* v. *lycopodium* and *C. cupressoides* f. *elegans* growing at 16 - 25 meters depth in the U.S. Virgin Islands achieved maximum growth rates when adequate light for photosyntheses was available for a daily light period greater than 10 h/day (Williams and Dennison 1990).

Figure 8. ***Caulerpa scalpelliformis*** **can be considered potentially invasive in U.S. waters.**

Caulerpa scalpelliformis

In 1994 *C. scalpelliformis* extended its natural range northward, into Botany Bay, New South Wales, Australia (Davis et al. 1997). Within two years of introduction *C. scalpelliformis* rapidly spread in some areas exceeding 55% cover, while sessile invertebrate cover was reduced by half (Davis et al. 1997). This species can be considered potenially invasive in U.S. waters due to its ability to survive in similiar climates to those present in the U.S.

Recognized forms and varieties (as listed in Guiry and Dhonncha 2004)

Caulerpa scalpelliformis var. *denticulata* (Decaisne) Weber-van Bosse
Caulerpa scalpelliformis var. *intermedia* Weber-van Bosse
Caulerpa scalpelliformis f. *dwarkensis* Børgesen
Caulerpa scalpelliformis f. *denticulata* (Decaisne) Svedelius
Caulerpa scalpelliformis f. *intermedia* (Weber-van Bosse) Svedelius

Regional distribution (as cited in Guiry and Dhonncha 2004)

Europe, Mediterranean Basin, Caribbean Basin, West Africa, East Asia, South-east Asia, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

Reproduction

Specimens of *C. scalpelliformis* collected off the Oki Islands, Japan were found to be dioecious, with separate plants containing male and female parts (Kajimura 1977). Gametes were observed to be formed not only in erect assimilators (fronds) and rhizomes, but also in the rhizoids (root tufts) [Kajimura 1977].

Caulerpa mexicana

The blades of *C. mexicana* are pinnately divided (each blade or leaf has several leaflets) with a flat midrib, pinnules (leaflets) are closely placed, sometimes overlapping, opposite, ascending, flat oval to oblong and basally narrowed to acuminate (Olsen et al. 1998).

Figure 9. ***Caulerpa mexicana*** **can grow to 15m deep.**

Recognized forms and variety (as listed in Guiry and Dhonncha 2004)
Caulerpa mexicana var. *pluriseriata* W.R. Taylor
Caulerpa mexicana f. *pectinata* (Kützing) W.R. Taylor
Caulerpa mexicana f. *vietnamica* Pham-Hoàng Hô
Caulerpa mexicana f. *laxior* (Weber-van Bosse) W.R. Taylor

Regional distribution (as cited in Guiry and Dhonncha 2004)
Europe, Mediterranean Basin, Caribbean Basin, West Africa, Eastern South America, South-east Asia, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

U.S. distribution
Florida Keys, southwest coast of Florida, Tampa Bay region (Walters 2004) and Hawaii (Peyton 2004)

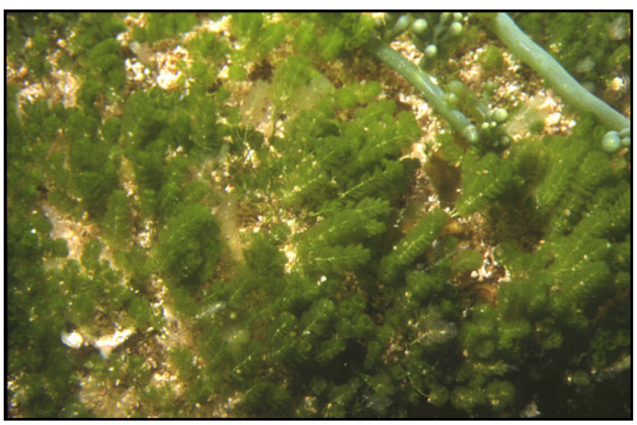
Environmental tolerance
C. mexicana has been observed growing at depths of ranging from 3 to 15 m (Johnston 1969).

Caulerpa verticillata

Caulerpa verticillata is another fast growing species of *Caulerpa*.

Recognized forms (as listed in Guiry and Dhonncha 2004)
Caulerpa verticillata f. *charoides* Weber-van Bosse
Caulerpa verticillata f. *acuta*

Figure 10. *Caulerpa verticillata* is another fast growing species of *Caulerpa*.



Regional distribution (as cited in Guiry and Dhonncha 2004)
Caribbean Basin, West Africa, Eastern South America, East Asia, South-east Asia, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

U.S. distribution
Florida Keys (Walters 2004), Guam and Hawaii (Peyton 2004)

Caulerpa sertulariodes

Recognized forms (as listed in Guiry and Dhonncha 2004)
Caulerpa sertularioides f. *longipes* (J. Agardh) Collins
Caulerpa sertularioides f. *brevipes* (J. Agardh) Svedelius
Caulerpa sertularioides f. *longiseta* (Bory de Saint-Vincent) Svedelius
Caulerpa sertularioides f. *corymbosa* W. R. Taylor
Caulerpa sertularioides f. *farlowii* (Weber-van Bosse) Børgesen
Caulerpa sertularioides f. *flagellata* (Weber-van Bosse) Weber-van Bosse
Caulerpa sertularioides f. *umbellata* (Weber-van Bosse) Svedelius

Regional distribution (as cited in Guiry and Dhonncha 2004)
Europe, Caribbean Basin, West Africa, Eastern South America, East Asia, Western North and Central America, South-east Asia, Western South America, Pacific Islands, East Africa, Indian Subcontinent and Indian Ocean, Australia and New Zealand.

U.S. distribution
Florida (Walters 2004), Oahu, Hawaii, Maui (Frisch 2003), Guam and Saipan (Peyton 2004).

Environmental tolerance
Stolon elongation rates for *C. sertularoides* growing in a deep water (16-25 m) sand canyon in the Caribbean were very fast, ranging from 0.93 to 1.92 cm/day (Williams et al. 1985).

Although this species may not be invasive in Florida and the Hawaiian Islands, it has the potential to be invasive in coastal California due to its rapid growth rate.

Figure 11. *Caulerpa sertularioides* has the potential to be invasive in coastal California.

2.3. Assessment of historic *Caulerpa* introductions

While it is difficult to accurately attribute historic introductions of non-native *Caulerpa* species to specific vectors there has been broad speculation as to likely vectors and pathways. In most historic cases dumping of *Caulerpa* from aquaria is suspected as the pathway for introduction of non-native species of *Caulerpa* (Meinesz 1999, Komatsu et al. 2003, SCCAT 2003a). *Caulerpa* species may be introduced into aquaria intentionally on unintentionally on "live rock" (Frish 2003 and Walters per. comm. 2004) or as an aquarium plant used to remove excess nitrogen from the water. One survey of California aquarium stores found that twelve species of *Caulerpa*, including *C. taxifolia*, were available for sale that are capable of surviving once introduced into temerpate waters (Frisch 2003). Once introduced, *Caulerpa* species can spread via fragmentation, facilitated by both natural and anthropogenic causes such as anchor fouling, fishing nets, ballast water and other vectors.

2.3.1. History of *Caulerpa taxifolia* introduction in California

In June 2000 divers detected *C. taxifolia* in the Agua Hedionda Lagoon located in Carlsbad, California (CA) (SCCAT 2003a). The publicity generated led to the discovery of a second population of *C. taxifolia* 75 miles north of Carlsbad, in Huntington Harbor (SCCAT 2003a). Genetic examination later determined that these colonies were of the same strain that is widespread in the Mediterranean (Jousson et al. 2000).

The SCCAT (Southern California *Caulerpa* Action Team) [2003a] provides a detailed description of the sites where *C. taxifolia* was found in 2000. At Agua Hedionda the *C. taxifolia* infestation, first discovered in spring 2000, covered 1,047 m^2. Later that summer, additional patches covering 308 m^2 were found east of the original Agua Hedionda site. At the Huntington Harbor site hundreds of small *C. taxifolia* colonies were spread over a 10,585 m^2 area, although the individual colonies were much less dense than at the Aqua Hedionda site.

These two infestations probably resulted from fragments that survived being dumped from a marine aquarium into a storm drain or directly into the water bodies (SCCAT 2003a). However it was introduced, *C. taxifolia* was able to survive, and to spread over large areas, primarily in eelgrass (*Zostera marina*) beds (SCCAT 2003a).

2.3.2. History of *Caulerpa brachypus* introduction in Florida

Lapointe (HBOI 2000) reports that since his group began to study *C. brachypus* in a deep-water reef off the shores of Riveria and Palm Beach, Florida its population has spread from a few small patches to cover acres of reef. The spread of *C. brachypus* has raised concerns because of its potential impact on the reef ecosystem off the southeastern coast of Florida (HBOI 2000). The rapid spread and abundant patches of *C. brachypus* are thought to be driven by the discharge of nutrient rich, secondarily treated sewage off the coast of Palm Beach, Florida (HBOI 2003).

Divers first discovered *C. brachypus* off the coast of southern Florida while conducting a study for the State of Florida Harmful Algal Bloom Task Force (Lapointe and Barile 2001). Subsequent research suggests that the population detected off the coast of Florida is actually *C. brachypus* var. *parvifolia* (Lapointe personal comm.).

2.3.3. History of *Caulerpa taxifolia* introductions in other regions

C. taxifolia has been cultivated in aquariums of Europe since the late 1960s (Meinesz 2002a). A particularly cold tolerant strain of *C. taxifolia*, the Mediterranean strain, was first noticed in the wild, off the Mediterranean coast, in 1984 (Meinesz 2002a). Since its detection *C. taxifolia* (Mediterranean strain) has steadily spread throughout the northwestern Mediterranean Sea colonizing over 130 km^2 (Meinesz 2002a).

Since it was first observed in the wild in the northwestern Mediterranean Sea, *C. taxifolia* has steadily expanded its range (Meinesz 2002a). In 2000 the Mediterranean strain of *C. taxifolia* was also found growing off the southern Australia coast (Millar 2002).

Since *C. taxifolia* was first identified in New South Wales (NSW) during April 2000 in Port Hacking, 30km south of Sydney, it has also been identified in seven other estuarine locations in Australia (NSWF 2004). While the origin of the NSW populations is unclear, the algae is likely to have been introduced from the aquarium industry, for example by cleaning tanks and disposing of unwanted algae into a waterway (NSWF 2004). The NSW Government has allocated $1 million per year to focus on a comprehensive control program for *C. taxifolia*. The program includes removing the algae from sale in the aquarium trade, controlling infestations in priority areas and eradicating them where possible, educating the public, and restricting the use of fishing gear to help stop the spread of the algae (NSWF 2004).

In 1992 and 1993 the Mediterranean strain of *C. taxifolia* became temporarily established in the Sea of Japan, probably due to aquarium dumping (Komatsu et al. 2003). The mean surface water temperature, for the coldest two months of the year in 1993, dropped to 9.8ºC which was lethal to the *Caulerpa* and prevented its successful establishment (Komatsu et al. 2003). A detailed literature review of *C. taxifolia* (Mediterranean strain) is presented in Appendix C.

<u>Dispersal Mechanisms</u>
Dispersal of *C. taxifolia*, within the Mediterranean, is thought to be caused by both natural and anthropogenic mechanisms. The spread of *C. taxifolia* is dependent on fragmentation and subsequent vegetative reproduction by fragments (Thibaut and Meinesz 2002). Fragmentation can occur by disturbances due to storms, invertebrates or other natural mechanisms leading to the spread of asexually produced clones over short distances (Meinesz 2002a). Boats and fishing nets are also suspected of spreading *Caulerpa* fragments over greater distances (Meinesz 2002a). One study found that 90% of *C. taxifolia* fragments spread a short distance and only 10% of fragments spread as far as a few hundred meters (Thibaut and Meinesz 2002).

C. taxifolia fragments do not normally float, but researchers have observed mucilaginous and filamentous algae overgrowing cold stressed *C. taxifolia* fronds in the Mediterranean, causing them to float (Chisholm et al. 2000). This buoyancy caused by algal overgrowth could provide a mechanism for long range dispersal (Chisholm et al. 2000). The suggestion that cold water can lead to range expansion of *C. taxifolia* is supported by the correlation between a fall in seawater temperature, in the Mediterranean, between 1989 and 1991 and the subsequent major outward spread of *C. taxifolia* (Chisholm et al. 2000).

Shelter provided by other resident plants can facilitate the establishment of *C. taxifolia* fragments (Ceccherelli 2002). Ceccherelli (2002) found that the spread of *C. taxifolia* and *C. racemosa* is strongly dependent on habitat type. Areas with diverse macroalgal assemblages composed of encrusting, turf and erect species were less likely to be successfully invaded by either *Caulerpa* species. In general the presence of dense, low-growing (turf type) species of macroalgae promoted the spread of both *C. taxifolia* and *C. racemosa* (Ceccherelli 2002). Findings suggest that the invasive character of *C. taxifolia* and *C. racemosa* species increase over time as more turf habitat is created by existing *Caulerpa* colonies (Ceccherelli 2002).

2.3.4 Eradication and control efforts for *Caulerpa taxifoli*a

<u>California</u>
The Southern California *Caulerpa* Action Team (SCCAT), a collaborative team of scientists, regulators, resource managers and stakeholders, was formed in June 2000 to respond to the *C. taxifolia* infestation. The infestation sites were surveyed and SCCAT decided to attempt an eradication of the introduced alga. Eradication methods were coordinated by the biological consulting firm that first detected the *C. taxifolia* colonies, Merkel & Associates of San Diego.

SCUBA divers covered the infested sites with 20 mil and 30 mil thick PVC tarps (Woodfield per. comm. 2004) and secured the edges of the tarps with sandbags. After the tarps were secured, divers pumped liquid chlorine or inserted solid chlorine pucks under the sealed tarps.

According to Anderson (2002) the following elements were considered essential to any successful *C. taxifolia* eradication attempt at the two infestation sites in California:

- Delineate the boundaries of the infestation
- Contain all plants to prevent immediate spread
- Kill and/or remove all live plants and propagules
- Evaluate the effectiveness of treatment and containment actions
- Prevent any reintroductions
- Monitor for both spread and reestablishment
- Implement an effective public awareness and education program
- Initiate new, appropriate, enforceable regulations
- Conduct appropriate, relevant and timely research
- Declare successful eradication only when no living parts are detected for three to five years following the last treatment

The synergy of elements surrounding the California introduction sites led to a strong rapid response. These elements, as described in Anderson (2002) include:

1) Political awareness
2) Political will
3) Scientific expertise
4) Fortuitous detection and location
5) Essential components of an eradication program were assembled through multi-agency consensus (SCCAT decisions and actions)
6) A field team was already in place
7) Treatment technologies were quickly evaluated and priorities were assigned
8) A group of dedicated people were committed to eradication
9) Funds and other resources were rapidly accessed **(*without this essential element the eradication would not have been possible**)

To stop the spread of *C. taxifolia*, stakeholders helped to initiate legislation to prohibit the sale of *C. taxifolia* and morphologically similar *Caulerpa* species in California. Other measures have been ongoing to monitor any spread and minimize the risk of reintroduction.

Divers with Merkel & Associates continue to survey the Aqua Hedionda and Huntington Beach sites for signs of *C. taxifolia* regeneration. Recreational divers have been asked to report any sightings to dive shops and to the California Department of Fish and Game. At Aqua Hedionda no new colonies have been found since fall of 2002. At Huntington Harbor no new colonies have been found since winter 2002. The current cover of *C. taxifolia* at both sites is estimated to be 0 m^2 (Woodfield and Merkel 2004).

While it is important to look on the SCCAT response as a successful response, the success of the SCCAT model must be deconstructed for general application. It is unlikely that the exact combination of elements that found foci in SCCAT would be present at all other locations in U.S. waters in which invasive *Caulerpa* species could establish.

Croatia

In Stari Grad Bay, off the coast of Croatia, the spread of *C. taxifolia* colonies has been less successful and eradication has been achieved in limited areas (Zuljevic and Antolic 2002a). Extraction of clonal colonies with suction pumps proved to be an efficient method for the removal of small colonies, up to one meter in diameter, however this method frequently leaves fragments that can regenerate. To be effective this method needs to be repeated after a short period of regeneration (Zuljevic and Antolic 2002a). Control of *C. taxifolia* has also been attempted by covering colonies with tarps of four meter wide, 0.15 millimeter thick black plastic tarps. This method was found to be useful for all types of substrata, but is limited by damage inflicted to the tarps by anchoring, fishing or storm events (Zuljevic and Antolic 2002a).

New South Wales, Australia

Since *C. taxifolia* was discovered in New South Wales, Australia in 2000, a variety of control and eradication attempts have been made. Millar (2002) has documented the methods being used to control *C. taxifolia* in New South Wales. Hand removal by divers of a four square meter *C. taxifolia* colony in fine sediments appeared to be successful, but the colony grew back within six months. Other methods such as mechanical removal with suction pumps, smothering with tarps and applying rock salt have shown mixed success and their effectiveness is still being evaluated (Millar 2002). Researchers from New South Wales Fisheries have thus far concluded that applying swimming pool salt to *C. taxifolia* patches is the most effective treatment method as it results in death of the weed colonies in a matter of hours (NSMF 2004). The concentration of pool salt need to be tailored to particular site conditions to be effective.

Biological Control Agents

Scientists have tested two Mediterranean sea slugs, *Oxynoe olivacea* and *Lobiger serradifalci*, in aquariums and the open ocean as potential biological control agents of *C. taxifolia* (Thibaut and Meinesz 2000). Scientists have also studied the possible use of two tropical sea slugs, *Elysia subornata* and *Oxynoe azuropunctata*, as biological control agents (Meinesz 2002b).

Subsequent aquarium studies have demonstrated that feeding by *Lobiger serradifalci* causes fragmentation of *C. taxifolia* fronds and most of the fragments are capable of regenerating into plants (Zuljevic et al. 2001). Thus *L. serradifalci* is not an appropriate control agent, and could contribute to the spread of *C. taxifolia*. The same study found that feeding, on *C. taxifolia*, by *Oxynoe olivacea* did not result in fragmentation of fronds.

2.4. Regulation of *Caulerpa* species in the U.S.

C. taxifolia (Mediterranean strain) was listed as a federal noxious weed, by the United States Department of Agriculture (USDA) under authority of the Plant Protection Act on March 16, 1999. This listing prohibits importation, entry, exportation, or movement in interstate commerce of this particular strain of *Caulerpa taxifolia*.

The California Fish and Game Code 2300 prohibits the sale, possession, importation, transport, transfer and release of nine species of *Caulerpa*: *C. ashmeadii*, *C. cupressoides*, *C. floridana*, *C. mexicana*, *C. racemosa*, *C. sertulariodes*, *C. scalpelliformis*, *C. taxifolia*, and *C. verticillata*. Possession of these nine *Caulerpa* species can be permitted for scientific research only. Any person who violates section 2300 is subject to a civil penalty of not less then $500 and not more then $10,000 for each violation.

Title 46 of the South Carolina Code of Laws, also known as the South Carolina Noxious Weed Act (SCNWA), places restrictions on *C. taxifolia*. The SCNWA provides far reaching powers to seize, quarantine, treat, destroy apply other remedial measures, to export, return to shipping point, or otherwise dispose of *C. taxifolia*. The SCNWA also includes provisions to deter persons from spreading nuisance aquatic weeds including fines not exceeding $500 and/or imprisonment not exceeding one year (SCDNR 2004).

Section 800: 20-3-2 of the Oklahoma Department of Wildlife Conservation code lists *C. taxifolia* as a declared noxious aquatic plant species (ODWC 2004).

At this time, no other U.S. state is known to restrict the sale or transport of any *Caulerpa* species.

See Appendix A "A Prevention Program for the Mediterranean Strain of *C. taxifolia*" for a list of other regulations that may apply to *Caulerpa* species.

3. Impacts of *Caulerpa* Introductions

The likely impacts of new introductions of invasive *Caulerpa* species into US waters are difficult to predict since the impacts of this genus have primarily been studied in different ecosystems such as the Mediterranean Sea. Introductions of *Caulerpa* species into waters of the U.S. have been controlled (*C. taxifolia* in California) or remain largely unstudied (*C. brachypus* and others in Florida), so the likely impacts to U.S. coastal marine ecosystems of *Caulerpa* introductions can not be certain. Still it is possible to make predictions of likely impacts based on documented impacts in other regions of the world.

3.1. Ecological Impacts of *Caulerpa* Introductions

In the Mediterranean Sea, large infestations of *C. taxifolia* have been shown to impact the biodiversity of many species of algae, benthic invertebrates and fishes (Meinesz 2002a, see Appendix C). Evidence from *Caulerpa* introductions into ecologically similar regions outside of the U.S. suggest potential impacts if *Caulerpa* species are introduced into waters of the U.S. to which they are not native.

Potential impacts of invasive *Caulerpa* introductions include:

- Competition with native plant species
- Alteration of predator-prey interactions (via habitat displacement and/or changes in camouflage requirements)
- Direct toxicity to herbivores
- Indirect toxicity to invertebrates due to degraded compounds
- Shading and smothering of coral reefs

3.1.1. Direct and indirect impacts on marine plants

Marine plants are an important part of the ecosystem providing food and shelter for resident species. Replacement of native marine plant species with invasive *Caulerpa* species could be expected to decrease important habitat unless the resident plant species are capable of successfully competing with *Caulerpa*.

In the Mediterranean Sea, the impact of *Caulerpa* on seagrass (*Posidonia oceanica*) communities has been studied. A study conducted from fall 1995 to fall 1996 found that *C. taxifolia* fragments were able to establish, in large numbers, at the margin of previously dominant sea grass (*Posidonia oceanica*) colonies in the Mediterranean Sea (Ceccherelli 2002). When both rock and dead *P. oceanica* mat substrates are present *C. racemosa* will produce more biomass on the mat, suggesting that mats are a more favorable substrate for overgrowth by *C. racemosa* than rock (Piazzi et al. 2001b). In the Mediterranean Sea the macroalgal community is impoverished when invaded by *C. racemosa*; species cover was reduced, the number and diversity of macroalgae fell, and the relative abundance among vegetation layers changed (Piazzi et al. 2001b). Competition with *C. taxifolia* and *C. racemosa* has been shown to decrease the longevity of leaves of *P.*

oceanica in the Mediterranean, possibly as a result of allelopathic compounds (Dumay et al. 2002b). Subsequently *P. oceanica* experiences high energy costs to replace leaves, which may lead to a shortage of nutrients necessary for growth (Dumay et al. 2002b). Other authors suggest that *Posidonia* may be able to effectively compete with *C. taxifolia* in areas where urban pollution is controlled, but that *C. taxifolia* will out-compete *Posidonia* in polluted areas (Jaubert et al. 1999). This suggestion is supported by the resilience of *Posidonia* beds to *C. taxifolia* invasion at depths as deep as 17m, in areas where urban wastewater receives secondary treatment prior to release (Jaubert et al. 1999).

C. prolifera has also been observed competing with other aquatic macrophytes by producing summer bloomss that shade out other species at the same time as water temperatures increase, causing the metabolic cost of vegetative maintenance for the other species to rise (Terrados and Ros 1991). One study showed that the suppression of *Cystoseira barbata* f. *aurantia* (an endemic macroalgae of the Mediterranean) by *C. taxifolia* varied seasonally, being the greatest during spring (Ferrer et al. 1997). Alternatively no effect was found on the growth of another macroalgae, *Gracilaria bursa-pastoris* when associated with *C. taxifolia* (Ferrer et al. 1997). In another study, chemical extracts from *C. taxifolia* inhibited the growth of *Cystoseira barbata* at lower concentrations than for the other algae, implying that the chemical extracts were producing the inhibitory effect observed on *Cystoseira barbata* (Ferrer et al. 1997). In Huntington Harbor, CA populations of a submerged aquatic plant (*Ruppia maritima*) were observed to be reduced where it grew with *C. taxifolia* compared to where it grew alone (Williams and Grosholz 2002).

Preliminary observations in the Mediterranean Sea suggest that *C. racemosa* also replaces native algae in invaded areas (Piazzi et al. 2001b). Observations of a new invasion demonstrated that *C. racemosa* had the largest impact on encrusting algal species, while erect algal species survived for at least 1 year after the invasion (Piazzi et al. 2001b). Since *C. racemosa* stolons have been observed to trap sediments, its presence may interfere with erect macroalgal species over time (Piazzi et al. 2001a). The rapid effects of competitive interactions between *C. racemosa* and other macroalgal species suggest direct interactions through allelochemical substances (Piazzi et al. 2003).

3.1.2. Direct impacts on marine herbivores

Species in the order Caulerpales contain many types of distasteful terpenoids and nitrogenous compounds. Of all the documented compounds caulerpin and caulerpenyne, also known as CYN, are the most studied in the genus *Caulerpa* (Lemee et al. 1993). The high degree of biological activity of CYN supports the hypothesis that this compound serves as a defense mechanism against herbivores and pathogens in tropical waters (Paul and Fenical 1986). In a broad study of nine species of *Caulerpa* from tropical regions of the Pacific and Atlantic, CYN was found to exist in concentrations of 40-50% of the organic extract and to constitute 1.0 to 1.5 % of dry weight (Paul and Fenical 1986). The compound CYN inhibits the growth of microorganisms, and the development of fertilized urchin eggs, and is toxic to the larval and adult stages of potential herbivores (Paul and Fenical 1986). Caulerpin, by comparison, shows little if any toxicity to herbivores (Paul and Fenical 1986). Paul and Hay (1986) recommend that concentrations of caulerpenyne, not caulerpin, should be considered when assessing the susceptibility of species and populations of *Caulerpa* to herbivores.

Caulerpenyne has demonstrated a variety of toxic effects in several organisms at different stages of their growth (Dumay et al. 2002). Laboratory tests indicate that various extracts from *C. taxifolia* can be toxic to mammals and sea urchin (*Paracentrotus lividus*) eggs (Lemee et al. 1993). Research indicates that CYN represents an ecological risk for microorganisms and multicellular animals that live close to *C. taxifolia* (Lemee et al. 1993). Table 1 shows the results of toxicity assays conducted by Paul and Fenical (1986) on common marine invertebrates and vertebrates, which demonstrated that CYN is toxic to various life stages of Pacific sea urchins (*Lytechinus pictus*) and two species of damselfish (*Pomacentrus coruleus* and *Dascyllus aruanus*). In addition, they found that CYN inhibited the growth of all fungi (5 species) and all bacteria (4 species) when CYN was applied to their colonies (Paul and Fenical 1986). The addition of CYN extracts to food deterred feeding by damselfish and juvenile conchs (*Strombus costatus*) and 50% of the conchs died after ingesting the CYN-tainted food (Paul and Fenical 1986).

Table 2. The results of bioassays conducted on the toxicity of caulerpenyne to different organisms for different exposure times. The minimum effective dose for 100% lethality is indicated by ED 100 (adapted from Paul and Fenical 1986).

Bioassays on Toxicity of Caulerpenyne to Marine Organisms		
Bioassay	**Organism**	**Lethal Concentration in (µg ml-1)**
Fertilized egg cytotoxicities ED100	*Lytechinus pictus,* Pacific sea urchin	8
Sperm toxicity (30 min) ED100	*Lytechinus pictus,* Pacific sea urchin	8
Larval toxicity (1h) ED100	*Lytechinus pictus,* Pacific sea urchin	8
Larval toxicity (24 h) ED100	*Lytechinus pictus,* Pacific sea urchin	2
Fish toxicity (1h)	*Pomacentrus coruleus* , damselfish	20
Fish toxicity (1h)	*Dascyllus aruanus* , black and white damselfish	20

The concentration of defensive compounds can vary greatly between *Caulerpa* species (Vest et al. 1983). A comparison of species in Florida found that *C. ashmeadii*, *C. paspaloides*, *C. sertularioides* and *C. racemosa* var. *uvifera* contained caulerpicin (caulerpenyne) in concentrations ranging from 0.03 to 0.09 mg/dry gram, while other species including *C. cupressoides* and *C. prolifera* did not (Vest et al. 1983). This finding is supported by a study, which found that extracts from *C. cupressoides* were lethal to goldfish (*Carassius auratus*) while extracts from both *C. racemosa* and *C. peltata* were only moderately toxic (De Lara-Isassi et al. 2000). The amount of CYN in a particular *Caulerpa* plant also varies seasonally, as a function of inter-plant competition levels and according to the particular *Caulerpa* species (Dumay et al. 2002). A field study of the CYN content in *C. taxifolia* showed that concentrations were highest in fall, lowest in spring and decreased as competition with other plants increased (Dumay et al. 2002). A comparison between species present in the Mediterranean also showed that *C. taxifolia* contained a mean CYN content that was 80 times higher then that found in *C. racemosa* (Dumay 2002).

3.1.3. Indirect impacts on marine invertebrates

The chemical defenses in *C. taxifolia* may have indirect impacts on resident invertebrates that do not feed on *Caulerpa*. Caulerpenyne is very unstable in sea water (Amade and Lemee 1998). Researchers have observed CYN samples, in seawater, degraded by 50% in 4 hours and 95% in 24 hours (Amade and Lemee 1998). However, unidentified toxic metabolites resulting from the degradation of CYN can still be toxic (Amade and Lemee 1998). The degradation products, which are constantly produced by *C. taxifolia*, are toxic to sea urchin (*Paracentrotus lividus*) eggs and may therefore influence the planktonic and larval recruitment (Amade and Lemee 1998) of at least this species.

A study conducted in the Cap-Martin (France) region found that the species richness of benthic invertebrates was slightly lower in *C. taxifolia* meadows at 6m depth compared to reference sites (Bellan-Santine et al. 1996). In addition, the number of benthic invertebrates was much reduced in *C. taxifolia* meadows at depths of 6m and 10m compared to reference sites (Bellan-Santine et al. 1996). In Agua Hedionda Lagoon, California significantly lower abundances of amphipods, isopods, and spirobids were associated with areas containing *C. taxifolia* compared to eel grass (*Zostera marina*) sites (Tippets 2002).

Seasonal variation in the concentration of CYN in *C. taxifolia* has also been linked to seasonal changes in feeding behavior by sea urchins (*Paracentrotus lividus*) and biomass of fouling organisms in the Mediterranean Sea (Amade and Lemee 1998).

In New South Wales, Australia a reef invasion by *C. scalpelliformis* was correlated with a significant decline in benthic cover by sessile invertebrates (Davis et al. 1997). Within two years coverage of sessile invertebrates (mostly sponges) was reduced to 23% compared to a reference site without any *C. scalpelliformis*, which averaged 65% cover by sessile invertebrates (Davis et al. 1997).

Caulerpa brachypus has produced large patches that overgrow sponges, hard corals, octocorals, and even other algal species off the coast of northern Palm Beach County (Lapointe and Barile 2001). Lapointe (HBOI 2000) reports that since his group began a study of *C. brachypus* in deep-water reefs off the shores of Riviera and Palm Beach, Florida the *C. brachypus* population has spread from a few small patches to cover acres of reef. The rapid spread of *C. brachypus* is likely to have a negative impact on the reef ecosystem off the southeastern coast of Florida (HBOI 2000).

C. prolifera has been shown to increase sedimentation rates in a coastal lagoon on the Iberian Peninsula (Terrados and Ros 1991), which would also be likely to indirectly impact benthic organisms.

3.1.4. Indirect impacts on marine vertebrates

One preliminary study in Cap Martin (France) found that there was a complex relationship between the introduced *C. taxifolia* (Mediterranean strain) and littoral fish communities (Francour et al. 1995). The study found that although species richness of littoral fish was lower in local sites infested by *C. taxifolia*, there was no detectable global decrease in species richness, indicating that other factors, such as intense fishing, could be more critical in influencing global species richness (Francour et al. 1995).

3.2. Economies that Could be Impacted by *Caulerpa* Infestations

While there are many anecdotal reports of decreases in fisheries and recreation due to the introduction and spread of *Caulerpa* in the Mediterranean, it has not yet been possible to establish definitive cause and effect relationships between the presence of *C. taxifolia* and reductions in fish stocks and recreational activities.

Impact of *Caulerpa* on Mediterranean Fisheries

The deserted underwater landscape caused by *C. taxifolia* has been reported to damage the SCUBA diving business in France. Managers of French Riviera dive clubs report that only beginning divers are willing to dive in local waters carpeted with *C. taxifolia* (Boudouresque 2002). Local fishermen also report decreased catches in areas infested by *C. taxifolia* (Boudouresque 2002).

U.S. Fisheries that Could be Impacted by *Caulerpa*: What is at Risk?

Large portions of the coastal waters of Alabama, California, Florida, Georgia, Louisiana, Mississippi, South Carolina and Texas have temperature regimes that rarely drop below 10ºC throughout the year (NOAA 2004). This temperature range is well within the temperature limits of *C. taxifolia* (Mediterranean strain) as documented in the laboratory (Komatsu et al. 1997, Meinesz 1999) and close to the suitable temperature range for tested wild strains of *C. taxifolia* (Chisholm et al. 2000). For example, the ocean surface water temparatures on the coast of Oregon and South Carolina are close to the range required for the establishment of C. taxifolia (Mediterranean strain). The surface water temperature in some areas, such as Port Orford (Oregon) and Charlestown (South Carolina) do not typically fall below 10°C (50°F) [NOAA 2004] and these areas may be sufficiently warm to allow establishment of at least *C. taxifolia* (Mediterranean strain).

Since *C. taxifolia* (Mediterranean strain) was first found in the Mediterranean in 1988, U.S. annual commercial landings[2] have increased in the aggregate with great variability in landings between U.S. regions (NMFS 2004). In Florida for instance commercial landings have steadily declined since 1988 (Figure 13), while in California commercial landings have fluctuated but appear to be more stable in the long term (Figure 14) [NMFS 2004].

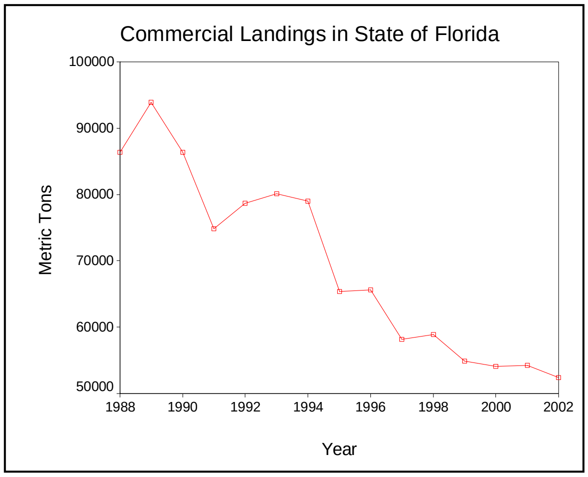


Figure 12. Commercial landings for Florida as reported by wholesale seafood buyers from 1988 to 2002 have steadily declined (data provided by NMFS 2004).

Even with some declines, the combined wholesale value of annual commercial landings in California and Florida alone averaged nearly $350 million over the last ten years, for which data was available (NMFS 2004). This figure does not include the many external benefits that are likely to be related to the wholesale seafood industry.

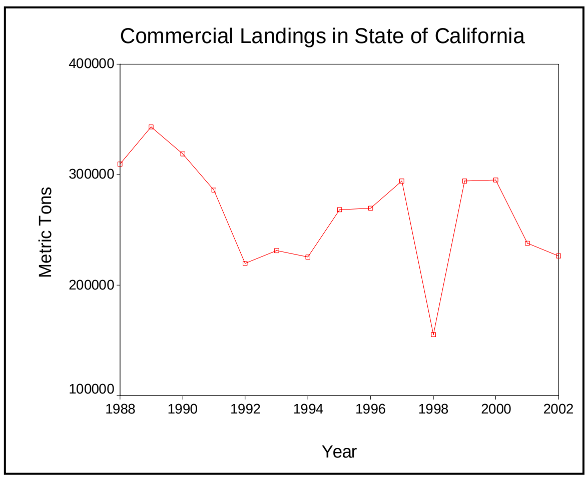


Figure 13. Commercial landings for California, as reported by wholesale seafood buyers for 1988 to 2002 have been highly variable (data provided by NMFS 2004).

Recreational fisheries could also be affected if a *Caulerpa* introduction were to establish and impact marine fishes. A survey of recreational fishers conducted in 2001 found that in California alone, the annual expenditures related to marine fishing exceeded $570 million (USDOI et al. 2001). A different survey conducted in 2000 estimated that total expenditure by residents in southern California related to marine angling was in excess of

[2] Figures are reported by wholesale seafood buyers and include primarily marine fish species, but also may include other marine species and plants.

$1.7 billion[3] (Gentner et al. 2001b). A similar survey conducted in the southeastern U.S. in 1999 estimated that total annual expenditure related to recreational marine angling exceeded $1.25 billion as displayed in Table 3 (Gentner et al. 2001a).

Clearly the commercial seafood industry and recreational marine fishing produce significant economic benefits in regions where non-native *Caulerpa* species have been able to establish populations (ex. *C. brachypus* in FL). While it is impossible to accurately predict the impact that non-native *Caulerpa* species could have on these industries, it has the potential to be significant.

State	Total Resident Expenditures (1999/2000 $)	Total Non-Resident Expenditures (1999/2000 $)
Florida	$7,832,619,000	$521,292,000
Georgia	$98,562,000	$5,034,000
Louisiana	$1,176,402,000	$29,636,000
Mississippi	$302,872,000	$10,079,000
North Carolina	$1,563,541,000	$237,087,000
South Carolina	$585,517,000	$51,923,000
Totals	**$1,162,794,000**	**$87,746,000**

Table 3. Total annual expenditures related to marine angling in the southeast U.S., estimated by survey, during 1999 and 2000, totaled over $1.1 billion for residents and over $87 million for non-residents (adapted from Gentner et al. 2001a).

3.3. Actual costs of *Caulerpa* control in the U.S.

3.3.1. Costs of eradication treatment in California

Eradication efforts targeting *C. taxifo*lia (Mediterranean strain) in California began in June 2000. Post-treatment monitoring of the sites is ongoing. The two sites had substantially different infestation characteristics that illustrate how costs can vary, as described below (Woodfield, personal communication 2004).

The cost of eradication is driven by surveillance and monitoring costs. To conduct a survey to detect patches as small as three meters wide, in an area the size of Huntington Harbor would cost in excess of $200,000 (Woodfield, per. comm. 2004).

In Huntington Harbor the infestation spread over approximately 8 acres. The total eradication operational cost in Huntington Harbor exceeds $1.2 million to date. Quarterly surveys that monitor for new *C. taxifolia* growth cost $15,000 for each survey (Woodfield per. comm. 2004).

In Agua Hedionda Lagoon the *C. taxifolia* infestation was relatively smaller, but monitoring was necessary over the larger Agua Hedionda area (approximately 200 acres). The total eradication treatment and monitoring costs, since June 2000, are approximately $2.5 million (Woodfield and Merkel 2004).

[3] The total expenditure on marine angling in all of California was estimated at $2.47 billion, but portions of the northern California coast drop below 10ºC for at least one month of the year and may be below the suitable temperature range for *Caulerpa taxifolia* (Mediterranean strain).

3.3.2. Costs of control and educational outreach in Florida

Florida has spent over $500,000 in direct allocations to research the impacts and spread of the non-native *Caulerpa* species in Florida (Lapointe, per. comm. 2004, and Walters, per comm. 2004).

4. Implementation Action Items

In February 2004, the *Caulerpa* Working Group agreed on goals and objectives of this National Management Plan as well as necessary actions to manage the genus *Caulerpa* in U.S. waters. The goals, objectives and actions that can achieve those objectives follow.

Goal 1: Prevent the introduction and spread of *Caulerpa* species to areas in U.S. waters where they are not native.

- Objective 1. Identify species of the genus *Caulerpa* that should be the focus of this national plan
- Objective 2. Analyze and prioritize pathways, vectors and foci that facilitate the introduction of species of the genus *Caulerpa* to U.S. waters and spread within U.S. waters
- Objective 3. Identify regulatory and policy needs and impediments to facilitate this goal

Goal 2: Early detect, rapidly respond to and monitor *Caulerpa* species in U.S. waters where they are not native.

- Objective 4. To develop further understanding of potential *Caulerpa* introduction sites and methods of detecting introduced populations of *Caulerpa* in U.S. waters
- Objective 5. To develop rapid response plans for newly detected *Caulerpa* introductions in different environments and to develop strategies and methods to control and manage populations

Goal 3: Eradicate *Caulerpa* populations, in waters to which they are not native, where feasible.

- Objective 6. To develop and evaluate strategies and protocols to eradicate *Caulerpa* species in U.S. waters where they are not native

Goal 4: Provide long-term adaptive management and mitigate impacts of populations of *Caulerpa* species in U.S. waters where they are not native and where eradication is not feasible.

- Objective 7. To develop site specific management plans for *Caulerpa* populations, where eradication is not feasible, in different environments and to develop strategies and methods to control and manage populations

Goal 5: Educate and inform the public, agencies and policymakers to advocate for preventing the introduction and spread of *Caulerpa* species.

- Objective 8. To develop education and outreach strategies for preventing and controlling *Caulerpa* introductions

Goal 6: Identify research needs and facilitate research to fill information gaps.

- Objective 9. To develop further understanding of potential and actual ecological and economic impacts of *Caulerpa* introductions

Goal 7: Review, assess progress and revise the management plan and continue developing information to meet national management plan goals.

- Objective 10. Provide guidance and coordination to governmental, non-governmental, and industry organizations in meeting these goals

4.1. Primary Priorities for Implementation

Table 4. Primary priority action items for the National Management Plan for the genus *Caulerpa*.

Primary Priority Action Items	
Goal	**Item**
Goal 1. Prevention	1-1) Work with aquarium industry on best management practices; labeling; outreach/education; more rigorous taxonomic identification; promote natives instead
	1-2) Work with aquarium industry to remove non-native *Caulerpa* from use
	1-3) Conduct a comprehensive pathways analysis & risk assessment for management
	1-4) Refine and publicize best management practices for handling *Caulerpa* samples
	1-5) Develop and post signage at high risk waterways
	1-6) Identify agencies involved in inspections and enforcement and analyze control gaps in the inspection process
Goal 2. Early Detection and Rapid Response	2-1) Decontamination and containment guidelines for infested areas to prevent spread
	2-2) Prioritize high risk sites for individual species
	2-3) Survey high risk sites regularly
	2-4) Create a National *Caulerpa* website
	2-5) Develop jurisdictional requirements for surveys prior to bottom disturbing activities
	2-6) Incorporate *Caulerpa* monitoring into other, existing, marine surveys-general plan specific
	2-7) Develop state and regional rapid response funds
	2-8) Designate and widely publicize state and regional contacts for *Caulerpa* sightings
	2-9) Develop an outreach kit that is ready for a rapid response situation
	2-10) Develop and refine detection methods
	2-11) Develop a national emergency response fund
Goal 3. Eradication	3-1) Develop and test strategies for treatment in high wave energy environments
	3-2) Develop a handbook of likely strategies for eradication in different environments
	3-3) Develop methods for assessing site specific efficacy of treatments
	3-4) Develop QA/QC methods for survey resolution/efficacy
	3-5) Evaluate the economic impacts of eradication
	3-6) Provide forum for stakeholder participation in eradication
Goal 4. Long-Term Management	4-1) Mapping delineation and extent of existing populations
	4-2) Create a *Caulerpa brachypus* management plan
	4-3) Develop regional guidance on how to integrate *Caulerpa* control with control of other algae
	4-4) Develop and implement scientific monitoring protocols of *Caulerpa* management
	4-5) Continue evaluation of biological control options
Goal 5. Education and Outreach	5-1) Develop rapid identification confirmation procedures
	5-2) Conduct outreach to customs brokers and importers, customs officers, agricultural inspectors, fishers, boaters, professional divers, recreational divers, dredge operators, agency staff etc
	5-3) Develop a display for Congress for Weed Awareness Week
	5-4) Create audience specific educational messages
	5-5) Conduct evaluation of outreach activities
	5-6) Develop and disseminate educational materials on identification of species
	5-7) Create outreach materials for clubs, hobbyists, etc.
	5-8) Provide outreach to aquarium service providers
	5-9) Develop a press kit
	5-10) Designate state/regional media contacts
	5-11) Conduct outreach about native *Caulerpa* species value

Primary Priority Action Items (continued)	
Goal	**Item**
Goal 6. Research Needs	6-1) Conduct research on managed populations
	6-2) Identify and research treatment options
	6-3) Research long range dispersal experiments and models
	6-4) Research natural/short range dispersal experiments and models
	6-5) Conduct research on the effect on biodiversity, alteration of reef food web; structure & function; fisheries
	6-6) Research environmental parameters where *Caulerpa* spp. may do well
	6-7) Conduct research on the ecological range prediction, of species thought to be invasive
	6-8) Research the reproductive potential and modes
	6-9) Conduct genetic research
	6-10) Research invasiveness and relationship to allelopathy and relationship to competition
	6-11) Research site parameters associated with establishment
	6-12) Develop scientific protocols and implement scientific monitoring of environmental conditions
Goal 7. Review & Assess Progress	7-1) Provide a forum for periodic review of management plan
	7-2) Facilitate review of site specific plans

<u>Action Items</u>

Prevention

1-1) Work with aquarium industry on best management practices; labeling; outreach/education; more rigorous taxonomic identification; promote natives instead; promote non-invasive ornamental alternatives (individual owners to large chains)
-Many authors have suggested that aquarium dumping probably led to the introduction and subsequent establishment of *C. taxifolia* into the Mediterranean and southern California. It has also been suggested that a 1997 introduction of *C. taxifolia* into the Sea of Japan was from aquarium dumping, but this did not result in successful establishment.

Educational outreach to the aquarium industry on identification of potentially harmful *Caulerpa* species, responsible labeling and benign alternative species could help to prevent any further introductions by aquarium dumping of *Caulerpa* species into waters of the U.S.

Alternatives to the use of potentially invasive *Caulerpa* species for ornamentation and nutrient scrubbing in aquariums should be identified and demonstrated. Demonstrations of better alternatives should be used to provide educational outreach to the aquarium industry and reduce the desire of consumers to use *Caulerpa* species in hobby and educational aquariums. Examples may include regionally native plants or artificial plants as appropriate.

One potential program for working with the aquarium industry is the Habitattitude campaign.

1-2) Work with aquarium industry to remove non-native *Caulerpa* from use (individual owners to large chains)
-Conducting educational outreach to organizations representing the aquarium industry on potentially harmful *Caulerpa* species may help to remove them from sale. The identification of profitable alternatives to the sale and use of *Caulerpa* would be more likely to prevent the use of non-native *Caulerpa*, particularly in areas where regulations do not already prohibit its sale. At a state level it may be possible to differentiate non-native from native *Caulerpa* species, but it would be less risky to discourage the use of any *Caulerpa* in aquariums and encourage the use of alternatives.

One potential program for working with the aquarium industry is the Habitattitude campaign.

1-3) Conduct a comprehensive pathways analysis & risk assessment for management (Anchor/chains, fishing gear, ballast water and other potential vectors)
-There is little information available about the potential for anchors, fishing gear, ballast water and other potential vectors to facilitate spread of *Caulerpa* species already present in U.S. waters to new areas and to introduce foreign species of *Caulerpa* to U.S. waters. Understanding the viability of these vectors is an important step in performing a risk assessment for the introduction and spread of *Caulerpa* species to new areas in U.S. waters.

1-4) Refine and publicize best management practices for handling *Caulerpa* samples (to prevent spread, for proper identification)
- After completion of action item 2-1 " Develop decontamination and containment guidelines for infested areas to prevent spread" the proper procedures for handling samples of potentially invasive *Caulerpa* species should be widely publicized, particularly in areas identified as high risk for introduction.

1-5) Develop and post signage at high risk waterways
-Informational signs should be posted near waterways at high risk for *Caulerpa* introductions. The signs should provide basic information about *Caulerpa*, its identification, sample handling techniques and contact information to report a sighting or send a sample to.

1-6) Identify agencies involved in inspections and enforcement and analyze control gaps in the inspection process
-Identification of different inspection agencies will allow for the distribution of specifically targeted educational materials. In addition to the identification of agencies, contact persons for particular inspection programs should be identified to facilitate the dissemination of information as action items are implemented.

A thorough analysis of the inspection process for high-risk cargo entering the U.S. or being transported within the U.S. should lead to the identification of regulatory and enforcement gaps in the current inspection process

Early Detection and Rapid Response

2-1) Develop decontamination and containment guidelines for infested areas to prevent spread (recreational user issues, anchors, jet ski, etc.)
-In order to prevent the spread of *Caulerpa* species once they have been introduced to U.S. waters, it is necessary to develop effective decontamination and containment guidelines to address a broad range of potential vectors and introduction sites. Development of decontamination techniques and analysis of containment guidelines should be refined in advance of new introductions to facilitate an effective rapid response to any new *Caulerpa* introductions.

2-2) Prioritize high risk sites for individual species (based on biology, physiological limitations, etc.)
-Once potentially invasive *Caulerpa* species have been identified, information on the limitations of those species combined with a comprehensive pathways analysis should then be used to identify and map marine areas in the U.S. that are at high risk for establishing *Caulerpa* populations.

Since aquarium releases (dumping) have been identified as likely vectors for *Caulerpa* introductions, proximity to residential areas, ease of access and aquarist access to *Caulerpa* species should be considered in risk assessments. (Because there are so many internet sites selling various species of *Caulerpa* it seems that aquarist access is a given.)

2-3) Survey high risk sites regularly

-A combination of agency staff and volunteer networks of divers should be actively organized to help in monitoring and early detection of any *Caulerpa* introductions. High risk areas, identified during implementation of action item 4-1, should be prioritized for monitoring with available resources.

2-4) Create a National *Caulerpa* website (GIS based database, tracking/eradication procedures and progress, clearinghouse for information)

-A National *Caulerpa* website should be created to serve as a clearinghouse for up-to-date information on *Caulerpa* issues in the U.S. To maximize exposure and use the website should be funded for the long term so that the internet address will remain the same over many years. The website should focus on all potentially invasive species of *Caulerpa* present in the U.S. (as determined by implementation of action item 4-1) and those likely to be introduced.

2-5) Develop jurisdictional requirements for surveys prior to bottom disturbing activities

-In high risk areas for the introduction of invasive *Caulerpa* species, the jurisdictional requirements should be researched and analyzed. The results of this analysis can be used to generate maps with information on the jurisdictional requirements for bottom disturbing activities for each high risk area. These analyses should be developed in coordination with agencies that have jurisdiction over the area of each analysis.

An example of this type of analysis is provided by the "*Caulerpa* Control Policy for Southern California" which was developed during the eradication response to *C. taxifolia* in southern California (http://swr.nmfs.noaa.gov/hcd/caulerpa/ccp.pdf).

2-6) Incorporate *Caulerpa* monitoring into other (existing) marine surveys

-Incorporating *Caulerpa* monitoring into existing surveys requires three elements:

1) Educational materials should be distributed to agency coastal marine monitoring programs informing them about the threats posed by *Caulerpa* species that are high risk in their areas.
2) Resource management agencies that conduct regular surveys of coastal marine areas should be encouraged to incorporate *Caulerpa* monitoring into their existing monitoring protocols.
3) Encourage Aquatic Nuisance Species Regional Panels to prioritize *Caulerpa* monitoring.

2-7) Develop state and regional rapid response funds

-States at high risk for introduction of *Caulerpa* should develop rapid response funds. States that share warm water coastlines, such as Texas, Louisiana, Mississippi, Alabama, Florida and Georgia should cooperate in development of such rapid response funds since the benefits of such a fund would cross jurisdictions. Special district authorities such as Aquatic Nuisance Species Regional Panels that coordinate interstate matters across these coast lines would be ideally suited to administer such a rapid response fund.

2-8) Designate and widely publicize state and regional contacts for *Caulerpa* sightings

- Once developed a contact person for each state as well as the state reporting procedures should be widely publicized, particularly in states identified as at high risk for the introduction of invasive *Caulerpa* species.

2-9) Develop an outreach kit that is ready for a rapid response situation
-General educational materials should be developed that can be rapidly deployed to any rapid response area in the U.S. Handouts and posters can be posted on the National *Caulerpa* website described in action item 2-4. Other educational materials such as informative signs and brochures should be available for distribution to rapid response coordinators in the U.S.

2-10) Develop and refine detection methods
-Research should be conducted to refine detection methods, to develop standardized monitoring protocols and to maximize the accuracy and efficiency of survey and detection methods. Detection and monitoring protocols should be refined for at least four likely scenarios:

1) incorporating *Caulerpa* monitoring into ongoing surveys of benthic marine areas,
2) monitoring sites once a new *Caulerpa* introduction has been detected,
3) monitoring populations undergoing eradication or management treatment and
4) monitoring areas after an eradication has been declared conditionally successful.

2-11) Develop a national emergency response fund of at least $10 million to complement state and regional funds.
- A national emergency response inter-agency fund should be developed for response to new *Caulerpa* introductions. Without adequate and available rapid response funds eradication of introduced *Caulerpa* populations is not feasible. The long term costs of control and the economic losses incurred by doing nothing are likely to exceed a one time cost of setting up an interest bearing emergency response annuity fund.

There are many ways to structure such a fund through collaboration between federal, state and tribal governments. If the fund was set up as an annuity then it would generate a revenue stream that could be used to address other aspects of *Caulerpa* management, if no new *Caulerpa* introductions are detected. Alternatively the fund could be set aside exclusively to address *Caulerpa* eradication and would grow annually in the absence of any new *Caulerpa* introductions.

However such an emergency response fund is structured, an inter-agency committee made up of scientists and agency staff should be established to determine the merits of appropriation requests to the fund. Nominations to the council should be made by representatives of funding agencies and from a pool of academic experts on marine invasions. All members of the council should be employed by agencies located in regions at high risk for *Caulerpa* introductions.

The fund council should be established so that the appropriation committee can be convened and make decisions on requests for emergency funds in a rapid time frame (14 days), once a new, potentially invasive, *Caulerpa* introduction has been confirmed via a standardized identification protocol. It will be essential for national coordinators of monitoring and early detection actions to have protocols established to notify and rapidly convene the *Caulerpa* emergency response fund council in the event of the detection of a potentially invasive *Caulerpa* population in U.S. waters.

Eradication

3-1) Develop and test strategies for treatment in high wave energy environments
It appears that treatment of the two sites containing *C. taxifolia* in southern California were successful, but notably they were both located in low energy environments. In a high wave energy environment, it is likely that *Caulerpa* would spread much more easily via fragmentation and treatment tarps might not stay in place. Treatment under a high wave energy environment would therefore necessitate containment and treatment considerations that were unnecessary in a low energy environment.

3-2) Develop a handbook of likely strategies for eradication in different environments
-A handbook of eradication strategies should be developed for different invasive *Caulerpa* species in a range of environments to which *Caulerpa* could be introduced. The handbook should be developed in part with input from members of the Southern California *Caulerpa* Action Team (SCCAT) on their eradication program for *C. taxifolia*. As information on eradication strategies develops the eradication handbook should be periodically updated.

3-3) Develop methods for assessing site specific efficacy of treatments
-Methods should be developed for assessing and monitoring the efficacy of *Caulerpa* treatments in a range of environments to which *Caulerpa* is likely to be introduced. These methods should include protocols for assessing effectiveness of treatments that have a minimal risk of allowing *Caulerpa* fragments to spread. The best available science and scientific approaches should be used determine the most efficacious treatments.

3-4) Develop quality assurance/quality control methods for survey resolution/efficacy
-Standardized methods to assess the actual resolution of survey methods should be developed and tested for accuracy. Methods should be developed for surveys ranging from broad population detection monitoring in high risk areas to fine resolution when confirming an eradication treatment. Methods should specify the most efficient methods based on the population status or introduction risk in a range of areas.

3-5) Evaluate the economic impacts of eradication (potential impacts of not eradicating, benefit cost analysis)
-A benefit cost analysis (BCA) should be conducted to evaluate the long-term costs and benefits of eradicating invasive *Caulerpa* species in U.S. coastal waters. This BCA should use the best financial, biological and economic information available from actual *Caulerpa* introductions to compare the long-term costs and benefits of invasive *Caulerpa* eradication or management vs. taking no action.

Quantifying the total economic costs of historic introductions and spread can help stakeholders to realistically assess the threat posed by additional introductions of invasive *Caulerpa* species. Summarizing this information can help to inform outreach educators about the likely economic cost of additional introductions of invasive *Caulerpa* species to various audiences.

3-6) Provide forum for stakeholder participation in eradication
-A standardized stakeholder forum process should be developed prior to the discovery of additional *Caulerpa* introductions. Having a process available in advance can save crucial time in implementing a rapid response.

Long-Term Management

4-1) Mapping delineation and extent of existing populations
-Existing populations of *Caulerpa* species in U.S. waters (including *C. brachypus*) should be mapped using global positioning system (GPS) devices and global information system (GIS) software. The U.S. *Caulerpa* populations should also be monitored at regular intervals to provide insights on density and distribution changes over time. This data should be freely available to Federal, State and local resource managers.

4-2) Create a *Caulerpa brachypus* management plan
-A management plan should be created for *C. brachypus* in coordination with or led by the State of Florida. The management plan should focus on reducing the abundance and controlling the spread of coastal population of *C. brachypus* and if possible moving toward eradication of this non-native *Caulerpa* species.

4-3) Develop regional guidance for integrating *Caulerpa* control with control of other algae
-Regional guidelines should be developed on how to integrate control of invasive *Caulerpa* species with the control of other algae on the basis of the best available science. These guidelines should be widely publicized and distributed to natural resource management agencies in regions at high risk for introduction of *Caulerpa* species. Aquatic Nuisance Species Regional Panels would be ideally suited to coordinate the developement of guidance for integrating *Caulerpa* control into other programs.

4-4) Develop and implement scientific monitoring protocols of *Caulerpa* management (effects on environment, ecosystem, economic impact, etc.)
-Science-based monitoring protocols should be developed and implemented in conjunction with active management of introduced *Caulerpa* populations to assess the economic, environmental and ecosystem impacts of active *Caulerpa* management. Documentation of associated impacts will help to inform future benefit cost analyses and guide policy makers on decision making in *Caulerpa* management.

4-5) Continue evaluation of biological control options
-The potential for effective use of biological agents for the control of introduced *Caulerpa* species should be researched particularly for existing populations, such as *C. brachypus*. A database of biological control options that are tested and the results of the tests should be maintained on a National *Caulerpa* website.

Research to date on biological control of *Caulerpa* species has focused on four sea slug species (*Oxynoe olivacea*, *Lobiger serradifalci*, *Elysia subornata* and *Oxynoe azuropunctata*). None of these slugs have proven to be a successful biological control for the Mediterranean strain of *C. taxifolia*, which is currently the most invasive *Caulerpa* species known. Other pathogens and herbivores should be researched for their ability to selectively control invasive species of *Caulerpa* and their ability to survive in waters identified as within the ecological range prediction of potentially invasive *Caulerpa* species.

Education and Outreach

5-1) Develop rapid identification confirmation procedures
-A procedure for coordinating the sampling and identification of potentially invasive *Caulerpa* species found in U.S. waters should be developed. The procedure should at the minimum consist

of a primary contact to report sightings as well as a list of certified identification experts who will be available for rapid identification as needed.

5-2) Conduct outreach to customs brokers and importers, customs officers, agricultural inspectors, fishers, boaters, professional divers, recreational divers, dredge operators, agency staff, K-12 etc.
-Educational outreach on the known and potential impacts of *Caulerpa* species should be conducted with individuals working and recreating in coastal marine areas such as fishers, boaters, divers, dredge operators and the staff of natural resource agencies. Educational materials can simultaneously inform these groups about state reporting procedures (as identified in action item 2-8 "Designate and widely publicize state and regional contacts for *Caulerpa* sightings").

Educational materials on invasive *Caulerpa* species should also be developed for customs and border inspectors based on an assessment of their needs and resources. Ongoing educational programs should educate inspection agents about the regulatory status of *Caulerpa* species and the likely vectors for their spread. Educational programs and materials should be regularly updated if regulatory status changes or new pathways are identified.

5-3) Develop a display for Congress for Weed Awareness Week
-A display that highlights issues related to invasive species of *Caulerpa* should be developed and presented at the National Invasive Weeds Awareness Week. Presenting up to date *Caulerpa* information at this event provides an opportunity to inform national lawmakers about the threats posed by *Caulerpa*.

5-4) Create audience specific educational messages (ex. pathways, users and regulators)
-A variety of educational outreach materials should be developed with messages tailored to specific audiences. Educational materials should focus on groups likely to be involved in introducing and controlling non-native *Caulerpa* species. Audiences likely to be vectors should be encouraged to eliminate behaviors that are likely to introduce and spread *Caulerpa* species. Audiences that are likely to be tasked with controlling introduced *Caulerpa* populations should be informed about the severe threat posed by some *Caulerpa* species and urged to rapidly respond to detected populations.

5-5) Conduct evaluation of outreach activities
-All outreach activities should be periodically assessed to determine if the desired information is being communicated effectively to the target audience and if that information is resulting in any actual behavior changes that lessen the likelihood of additional *Caulerpa* introductions. All evaluations of outreach activities should be analyzed to provide recommendations for improving effectiveness at achieving the desired behavioral changes. The ANSTF should require evaluation of outreach activities to be a priority item for regional panels in areas with a high potential for introduction and establishment of *Caulerpa*.

5-6) Develop and disseminate educational materials on identification of species
-Educational materials should be developed that provide identification information on potentially invasive *Caulerpa* species. These materials can link to the identification website referred to in action item 2-4 "Create a National *Caulerpa* website" and they can be periodically updated. Identification materials should be distributed to recreational divers, natural resource managers in coastal areas and other stakeholders likely to be the first to find introduced *Caulerpa* species in coastal waters of the U.S.

5-7) Create outreach materials for aquarium clubs, hobbyists, etc.
-Educational outreach materials should be developed with prevention messages targeting saltwater aquarium hobbyists. Education to this group should focus on the dangers posed by *Caulerpa* introductions to natural systems and the availability of alternatives to *Caulerpa* as an ornamental aquarium element.

A program, which shares goals similar to the Habittattitude campaign, is an ideal model for outreach. Habitattitude™ is about consumer awareness and responsible behaviors. By drawing attention to the potential environmental ramifications of the aquarium and water garden hobbies while promoting responsible consumer behaviors, Habitattitude™ avoids the definition debate surrounding "invasive species". Ultimately, the campaign seeks to eliminate the transfer and survival of any species outside of your enclosed, artificial system, which it has the potential to cause the loss or decline of native plants and animals (www.habitattitude.net/).

5-8) Outreach to aquarium service providers
-Educational outreach to aquarium service providers should be developed with a similar message as in action item 5-7. One key difference is that the outreach to service providers should focus on the profitability of using alternatives to *Caulerpa* as an ornamental aquarium element. Similarly to 5-7, this action item may use the goals of the Habitattitude campaign as a model for outreach.

5-9) Develop a press kit (media oriented sound bites for public affairs personnel [videos/pictures that are available], can link to site that has pictures, etc.
-A webpage that displays general mass media information on invasive *Caulerpa* species should be developed in conjunction with the National *Caulerpa* website described in action item 2-4. A set of high quality videos, pictures and concise information on *Caulerpa* species should be available for complementary download and use by media distribution organizations.

5-10) Designate state/regional media contacts [joint information center -federal/state/local]
-State and regional media contacts should be designated who can coordinate with the state identification contacts described in action item 2-8 "Designate and widely publicize state and regional contacts for *Caulerpa* sightings".

5-11) Conduct outreach about native *Caulerpa* species value
-In areas of the U.S. that contain native or cryptogenic *Caulerpa* species all educational outreach materials should clearly differentiate species that are not harmful from introduced invasive *Caulerpa* species. It may also be necessary to conduct outreach to divers about identification and value of native *Caulerpa* species to prevent unnecessary destruction of native *Caulerpa* species. Prior to conducting outreach about valuable *Caulerpa* species, it is crucial to have clear scientific evidence to support the role of individual *Caulerpa* species in the local ecosystem.as well as the species' region of origin.

Research Needs

6-1) Conduct research on managed *Caulerpa* populations
-Managed populations offer a unique opportunity to study *Caulerpa* species in a natural environment without risking the introduction of an invasive species. The biological, ecological and physical impacts of managed populations *in situ* should be studied to enhance understanding about the likely impacts of additional introductions of invasive *Caulerpa* species.

6-2) Identify and research treatment options
-A wide range of methods for extirpating and managing invasive *Caulerpa* species that are likely to be introduced into U.S. waters, should be tested. The best treatment options, for invasive *Caulerpa* species, should be determined by their effectiveness, rapidity of action and occurrence of the lowest possible incidental toxicity to non-target organisms. A database of failed or rejected approaches should be maintained on the National *Caulerpa* website described in action item 2-4.

6-3) Research long-range dispersal experiments and models
-The ability of invasive *Caulerpa* species to disperse over long distances and the vectors that may aid that dispersal are not well understood. Vectors that can lead to the unintentional long range spread of *Caulerpa* species should be modeled to determine the risk involved with particular vectors.

6-4) Research natural/short range dispersal experiments and models
-The vectors that can lead to short range dispersal of invasive *Caulerpa* species, once introduced to U.S. waters, are not well understood. Clarification on potential vectors that can disperse invasive *Caulerpa* over short distances would help to efficiently control spread of existing and potential future introductions in a rapid response or long term management situation. Researching dispersal of native *Caulerpa* species may provide a low risk method to model the spread of invasive *Caulerpa* species.

6-5) Conduct research on the effects on biodiversity, alteration of reef food web; structure & function; fisheries
-The impacts that invasive species of *Caulerpa* can have on biodiversity and ecosystems in coastal U.S. waters has not been scientifically researched. Projections of the ecological impacts of invasive *Caulerpa* introductions are derived from research conducted in the Mediterranean and anecdotal observations.

The introduced population of *C. brachypus* can be studied in a wild situation off the coast of eastern Florida to determine its impacts on the natural ecosystem of coastal eastern Florida. Laboratory studies could also be performed to study the likely impacts of other potentially invasive species such as *C. taxifolia*, *C. racemosa*, *C. prolifera*, *C. cupressoides*, *C. scalpelliformis*, *C. mexicana*, *C. verticillata* and others yet to be identified as potentially invasive.

6-6) Research environmental parameters where *Caulerpa* spp. may do well (disturbance regimes, light requirements, temperature, nutrients, salinity, herbivory, depth, flow, competitors, substrate)
-Limited research findings are available on the habitat requirements for *C. taxifolia*, but not for most potentially invasive *Caulerpa* species. A detailed list of the habitat requirements and environmental tolerance of all *Caulerpa* species would greatly inform risk analyses for individual species.

Limited research has suggested that certain types of environmental disturbances, such as nutrient pollution, can support invasion by *Caulerpa* species. More research on the interaction between environmental conditions and invasive characteristics of *Caulerpa* populations will help to focus monitoring and control of invasive *Caulerpa* species.

The light requirements of invasive *Caulerpa* species are not well understood. A better understanding of the light requirements and the relationship between the optical properties of the water column and invasiveness of *Caulerpa* species will inform risk analyses and prevention programs

6-7) Conduct research on ecological range prediction, of *Caulerpa* species thought to be invasive
-As knowledge of the habitat requirements of potentially invasive *Caulerpa* species increases, the information should be synthesized to make potential ecological range predictions for individual species.

6-8) Research reproductive potential and modes
-More research is needed on the reproductive potential and modes of potentially invasive *Caulerpa* species. Research should focus on the reproductive potential and modes under environmental conditions that mimic actual U.S. coastal conditions.

6-9) Conduct genetic research (relationship between invasive strains, genetic proximity of native strains, genetic relationship to invasiveness, etc.)
-Research on the genotypes of invasive species may reveal markers that could be used to predict invasive potential in other *Caulerpa* species. Aspects such as the genetic similarity between invasive strains, the similarity between invasive and native strains and the relationship of genetic characteristics to invasiveness should be researched. The results of genetic research could enhance the ability to predict which *Caulerpa* species could be invasive in U.S. waters.

6-10) Research invasiveness and relationship to allelopathy and relationship to competition
-The role that allelopathy may play in the ability of invasive *Caulerpa* species to out-compete native marine plants is not well understood. Research should be conducted to determine what if any chemical compounds produced by invasive *Caulerpa* species exert allelopathic effects on existing, marine plant communities.

6-11) Research site parameters associated with establishment (basic boundaries, physical and biological variables that contribute to establishment, etc.)
-Site parameters and other physical and biological variables that enable the establishment of invasive *Caulerpa* species remain largely undefined. A better understanding of the cultural requirements necessary for the establishment of non-native *Caulerpa* species will help to inform risk analyses and allow for efficient focus of necessary prevention programs.

6-12) Develop scientific protocols and implement scientific monitoring of environmental conditions (determine physical and biological impacts of *Caulerpa*)
-Where populations of invasive *Caulerpa* species exist in U.S. waters their physical and biological impacts on the surrounding environment and ecosystem should be studied to determine the breadth and extent of impacts. As information on the impacts of invasive *Caulerpa* species increases, the measurement of impacts should be standardized in protocols tied to key physical and biological parameters that can be monitored over time.

Review & Assess Progress

7-1) Provide a forum for periodic review of this National Management Plan
-In order for this National Management Plan (NMP) for the Genus *Caulerpa* to remain effective it will be necessary to reconvene the *Caulerpa* Working Group at least annually. However, if new introduced populations are discovered, then the CWG should convene more frequently to assess progress on plan implementation, to discuss new scientific and economic findings and their implications for the NMP, and to determine if the NMP requires updating.

Implementation of this NMP will be coordinated by the U.S. Fish and Wildlife Service Aquatic Nuisance Species Program with oversight from the Aquatic Nuisance Species Taskforce.

7-2) Facilitate review of site specific plans (some experts listed in Appendix B)
-Appendix B in this NMP lists CWG members as well as other experts who may be able to facilitate review of site specific plans for the eradication and management of non-native invasive *Caulerpa* species.

4.2. Secondary Priorities for Implementation

Table 5. Secondary priority action items for the National Management Plan for the genus *Caulerpa*.

Secondary Priority Action Items	
Goal	Item
Goal 1. Prevention	1-7) Identify "look-alike" species
	1-8) Identify and address domestic sources of *Caulerpa* that may be used in aquaria
	1-9) Conduct comprehensive identification of locations in the world where invasive *Caulerpa* species may be shipped from
	1-10) Analyze the labeling of *Caulerpa* shipments – authority gap
	1-11) Encourage states/territories to evaluate and regulate non-native *Caulerpa* species
	1-12) Improve enforcement of internet shipments
	2-12) Develop pest alarm drill
Goal 4. Long-Term Management	4-6) Develop regional guidance on how to integrate *Caulerpa* control with non-algal species control
Goal 5. Education and Outreach	5-12) Develop and disseminate educational materials about scientific uncertainty related to *Caulerpa* management
	5-13) Identify/develop common names for *Caulerpa* species
Goal 6. Research Needs	6-13) Identify and research new pharmacological biocides/algacides
	6-14) Research deep water (beyond diver depth) survey methods
	6-15) Test models of site conditions for identified sp.
	6-16) Research hybridization between native and invasive strains
	6-17) Develop and refine remote sensing techniques

<u>Action Items</u>

<u>Prevention</u>

1-7) Identify "look-alike" species [final list outside of NMP].
-Species of *Caulerpa* can manifest variable morphology depending on a variety of environmental conditions that may exist within tolerable habitat. Inconsistent morphology can make identification of harmful *Caulerpa* species and differentiation of benign *Caulerpa* species difficult or impossible under any inspection scenario. Due to the difficulty with species level identification it is necessary to develop a comprehensive list of *Caulerpa* species with morphological characteristics similar to those of potentially harmful *Caulerpa* species. A list of look-alike *Caulerpa* species could be used to generate training materials for inspectors allowing for efficient and effective inspections to minimize the introduction risk for potentially harmful *Caulerpa* species. To establish definitive identification of species it may be necessary to build a genetic library of *Caulerpa* species as resources permit.

1-8) Identify and address domestic sources of *Caulerpa* that may be used in aquaria
-The distribution of native and introduced *Caulerpa* species within U.S. waters is not well understood. Any species presently in U.S. waters could potentially be harvested and distributed via the aquarium industry. While some *Caulerpa* species are endemic to parts of the U.S., they may still pose a threat to other areas of the U.S. to which they could be introduced through commerce. As a first step in the management of domestic sources of *Caulerpa*, it is necessary to identify and locate all *Caulerpa* populations present in U.S. waters. Once the *Caulerpa* populations are identified their spread to new areas via the aquarium trade and other potential vectors should be discouraged through public awareness campaigns and state regulations.

1-9) Conduct comprehensive identification of locations in the world where invasive *Caulerpa* species may be shipped from to the U.S.
-As available information on invasive *Caulerpa* species improves it will be possible to generate lists of locations from which invasive *Caulerpa* species might be shipped to the U.S., from global sources. This information would allow cargo inspectors to target high-risk imports such as identified vectors (ex. "live rock") originating from locations known to contain invasive species of *Caulerpa* (ex. the Mediterranean Sea).

1-10) Analyze the labeling of *Caulerpa* shipments – authority gap
-The labeling of *Caulerpa* species that are intentionally or incidentally imported should be researched and analyzed. Any significant authority gaps should be identified and recommendations for closing those gaps should be made to appropriate regulatory agencies or to the U.S. Congress, depending on whether the gap is regulatory or statutory.

1-11) Encourage states/territories to evaluate and regulate non-native *Caulerpa* spp. (detailed)
-States and territories should be informed about *Caulerpa* species that are potentially invasive and that may be imported to or originate from their jurisdiction. Several states including California, Texas, Louisiana, Mississippi, Alabama, Florida and Georgia border relatively warm coastal waters that are likely to support some *Caulerpa* species. Of these states, only California currently regulates the sale or movement of any *Caulerpa* species.

Oregon, South Carolina and North Carolina also have coastal water temperatures that may be moderately habitable to at least the Mediterranean strain of *C. taxifolia* and possibly other *Caulerpa* species. None of these states currently regulate any species of *Caulerpa*.

Because of the tropical locations of some U.S. Territories, they may have special issues in regulating non-native and native *Caulerpa* species and should be informed about the dangers that some tropical species of *Caulerpa* may pose if introduced to temperate waters.

1-12) Improve enforcement of internet shipments
-Special agents should continue to assess compliance with legal restrictions on the trade in the Mediterranean strain of *C. taxifolia* in commerce or trade conducted over the internet. Stakeholders should report evidence of trade in the Mediterranean strain of *C. taxifolia* in commerce conducted over the internet to the US Department of Agriculture Animal and Plant Health Inspection Service, Agricultural Internet Monitoring System. If additional *Caulerpa* species are placed on the federal noxious weed list, then federal agents should improve enforcement of trade restrictions of those species.

Multi-state groups could also form memoranda of understanding to aid in restricting the sale of restricted *Caulerpa* species to individuals in states which ban those species.

Early Detection and Rapid Response

2-12) Develop pest alarm drill (conducted periodically to assess ability to react)
- Once an emergency fund has been structured (action item 2-11"Develop a national emergency response fund of at least $10 million to complement state and regional funds"), the ability of the fund council to respond to the detection of a new *Caulerpa* introduction should be periodically tested (at least once a year). The alarm drill should assess the commitment of council members in

their roles by determining if the council could be rapidly convened on short notice and make a decision in less than 14 days.

The alarm drill should also test the ability of coordinators to assemble information necessary for eradication and to rapidly distribute that information to the appropriate action agency. A standardized eradication kit that can be distributed to action agencies in the event of an introduction should be developed prior to any alarm drills. This kit should include information on essential steps in any eradication process.

Long-Term Management

4-6) Develop regional guidance on how to integrate *Caulerpa* control with non-algal species control
- Regional guidelines should be developed on how to integrate control of invasive *Caulerpa* species with the control of non-algal species on the basis of the best available science. These guidelines should be widely publicized and distributed to natural resource management agencies in regions at high risk for introduction of *Caulerpa* species. Advances in understanding resulting from action item 6-7, "Conduct research on ecological range prediction, of *Caulerpa* species thought to be invasive" would facilitate this action item.

5-12) Develop and disseminate educational materials about scientific uncertainty relative to *Caulerpa* management (for agency staff, public, university extension programs etc.)
-Educational materials targeting agency staff, extension programs and academia should focus on the severity of threats posed by non-native *Caulerpa* species even though threats may not be quantified for all potential introduction areas.

Education and Outreach

5-13) Identify/develop common names for *Caulerpa* species
-Developing and disseminating common names for invasive *Caulerpa* species may make educational messages more accessible to more general audiences. Since most species of *Caulerpa* do not have common names this exercise would help to track commerce in *Caulerpa* and to disseminate information to the public about *Caulerpa* species.

Research Needs

6-13) Identify and research new pharmacological biocides/algacides (ex. may target a particular protein)
- Biocides and algacides for extirpation of invasive *Caulerpa* species that are likely to be introduced into U.S. waters should be researched with focus on finding the most effective, rapid acting compounds with the lowest incidental toxicity to non-target organisms. A database of failed or rejected approaches should be maintained on a National *Caulerpa* website, described in action item 2-4.

6-14) Research deep water (beyond diver depth) survey research (source/sink populations)
-Since many *Caulerpa* species are capable of surviving in relatively low light conditions and in deep coastal waters (>50m) beyond safe diving depths, source populations could exist without being detected by divers. Alternative methods should be developed to survey deep locations. Once methods have been developed, they should be compared to determine the most reliable and efficient methods.

6-15) Test models of site conditions for identified species
-As individual *Caulerpa* species are identified as potentially invasive they should be tested for their ability to survive and reproduce in site conditions that mimic those found in U.S. coastal waters. Great caution should be taken when conducting any such tests to assure that there is no chance for an accidental escape and introduction of the *Caulerpa* species being tested.

6-16) Research hybridization between native and invasive strains
-Potential and actual hybridization between native and invasive strains of *Caulerpa* should be researched to understand the potential impact of hybridization on native populations and if native *Caulerpa* species may enhance the fitness of invasive non-native *Caulerpa* species.

6-17) Develop and refine remote sensing techniques
-Efficient remote sensing techniques are essential for the efficient monitoring of *Caulerpa* populations and in large, high risk wild marine areas. Great progress has been made recently in underwater remote sensing technology and techniques. Refining the protocols for using underwater remote sensing to detect new *Caulerpa* populations would increase the chances of detecting populations early enough to eradicate.

4.3. Tertiary Priorities for Implementation

Table 6. Tertiary priority action items for the National Management Plan for the genus *Caulerpa*.

Tertiary Priority Action Items	
Goal	Item
Goal 1. Prevention	1-13) Identify existing assessments and summarize findings
	1-14) Research protection of coastal waters from nutrient pollution that favors *Caulerpa* outbreaks
	1-15) Use the Federal Noxious Weed list for regulating additional non-native *Caulerpa* species
	1-16) Evaluate economic incentives/penalties that can reduce risk of introductions
	1-17) Evaluate regulation of aquarium service industry
	1-18) Evaluate information strategies vs. regulatory enforcement results
	1-19) Develop guidelines for labeling high risk items

Action items

Prevention

1-13) Identifying existing assessments and summarize findings
-The vast world-wide distribution of *Caulerpa* makes it likely that many detailed assessments of the invasive characteristics of particular species may exist, probably in multiple languages. It would be much more efficient to identify and summarize existing assessments than to begin primary research on all of the species in such a large and diverse genus. A comprehensive identification and summarization or findings on *Caulerpa* species would allow organization to focus control policies on a more manageable subset of the total genus.

It is likely that this summary of existing assessments would facilitate effective implementation of many other action items, particularly those focused on educational outreach. However, this action item may be very difficult, time consuming and expensive to complete with no certain benefits. For these reasons this potentially essential action item was not ranked as a high priority.

1-14) Research protection of coastal waters from nutrient pollution that favors *Caulerpa* outbreaks (coordinate with agencies that have authority over pollution emission -EPA, NPS, etc.)
-Several studies have suggested that nutrient pollution may provide *Caulerpa* species with a competitive advantage over other marine plants and algae. Compliance with a host of environmental Acts and the policy direction provided by Executive Order 13112[4] should be considered when agencies are permitting the discharge of nutrient pollutants. Agencies may not routinely account for the effect of nutrient discharge on invasive species of *Caulerpa* when permitting discharges or analyzing policy alternatives. Research on the effects of coastal nutrient pollution should be coordinated with agencies such as the U.S. Environmental Protection Agency, which has authority over the approval of National Pollutant Discharge Elimination System (NPDES) permits.

1-15) Use the Federal Noxious Weed list for regulating additional non-native *Caulerpa* species
-A petition to list *C. taxifolia* (whole species) and *Caulerpa* (whole genus) was submitted to the USDA APHIS on July 6, 2003. The outcome of that petition should be determined before devoting resources to this action item. Currently the USDA APHIS is working on a pest risk assessment for the genus *Caulerpa* and will consider listing as Federal noxious weeds species of

[4] Federal Register/Vol. 64, No. 25/ Monday, February 8, 1999/ Presidential Documents.

Caulerpa that are determined to pose a risk to the United States and that meet the International Plant Protection Convention definition of a quarantine pest.

1-16) Evaluate economic incentives/penalties that can reduce risk of introductions (reward for reporting dumping, place to return unwanted plants, etc.)

-A detailed policy analysis of economically efficient incentives and penalties likely to reduce or eliminate introductions of invasive *Caulerpa* species should be completed for regions at risk for *Caulerpa* introductions. Such an analysis would inform local, state and federal lawmakers of the benefits and costs of such policies allowing for better decision making.

1-17) Evaluate regulation of aquarium service industry (in various states)

-An evaluation of the potential for *Caulerpa* introductions posed by the aquarium service industry in different states would allow state regulators to assess the risks posed by the industry in their state and by neighboring states that share borders with coastal areas.

1-18) Evaluate information strategies vs. regulatory enforcement results

-A detailed analysis of the effectiveness of information strategies and regulatory enforcement in preventing the introduction and spread of *Caulerpa* species in the U.S. should be conducted to determine what methods are the most efficient at preventing the spread of *Caulerpa* species. Specific information on the effectiveness of programs can help appropriators to efficiently allocate limited resources to the best use. An example would be a comparison of a voluntary program such as the Habitattitude campaign with regulatory enforcement to determine if either yielded greater results.

1-19) Develop guidelines for labeling high-risk items (flow chart)

-Many importers may not realize that they are moving items at high risk when transporting *Caulerpa* species. A flow chart providing guidance for labeling of items at high risk for intentional and incidental transportation of *Caulerpa* species should be developed. The guidelines should be provided to importers likely to transport high risk items.

4.4. Implementation Table for the National Management Plan for the Genus Caulerpa Action Items

Prioritized action items to be implemented are summarized in tabular form and detailed in narrative form beginning in Section 4.1 and continuing through Section 4.3.

Actions Funded in Thousands of Dollars					
Related Goal	**Action**	**Funded by**	**Implemented by**	**2004**	**2005**
Goal 1. Prevention	Education and outreach to aquarists and inspectors throughout the U.S. (focus on CA & FL)	USFWS, CSG, UCF	CSG, UCF		$27
Goal 4. Long-Term Management	*C. brachypus* initial risk assessment	NOAA, NMFS	IC	$20	
	Mapping extent and impacts of *C. brachypus* in FL	FL	HBOI	$500	
	National Management Plan for *Caulerpa*/CWG Coordination	USFWS	USFWS	$22	$20
Goal 5. Education and Outreach	*Caulerpa* educational outreach in FL	TBEP	UT	$7	
	Survey of *Caulerpa* in FL and educational outreach	PCEF	UCF	$31	
	Develop rapid ID an confirmation procedures	USFWS, FDACS	FDACS		$30
	Create a National *Caulerpa* website	USFWS, UAZ	UAZ		$19

Abbreviation	Organization
CSG	California Sea Grant
FDACS	Florida Department of Agriculture and Consumer Services
IC	Independent Contractor
NMFS	National Marine Fisheries Service
NOAA	National Oceanic and Atmospheric Administration
PCEF	Pinnellas County Environmental Fund
TBEP	Tampa Bay Estuary Program
UCF	University of Central Florida
UT	University of Tampa

Related Actions Funded:

- Over the period of 1999 through 2002 the U.S. Fish and Wildlife Service spent $83,800 on prevention planning and action planning for *Caulerpa taxifolia* (Mediterranean Strain).
- Over the period of 2000 through 2003 a broad coalition of federal, state and local organizations spent over $3,700,000 on rapid response, eradication, monitoring and educational outreach in response to the infestations of *Caulerpa taxifolia* (Meditarranean Strain) found in two Californian sites. Many of these activities continue through 2005.

5. References Cited

Algalbase. 2004.
http://www.algaebase.org/
Accessed by David Bergendorf (USFWS) February 13-August 31, 2004.

Amade, P. and Lemee, R. 1998. Chemical defence [defense] of the mediterranean [Mediterranean] alga *Caulerpa taxifolia*: variations in caulerpenyne production. Aquatic toxicology 43: 287-300.

Anderson, Lars W.J. 2002. *Caulerpa taxifolia* in the United States: Rapid response and eradication program. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Aquatic Nuisance Species Taskforce (ANSTF). 2002. Spring Meeting February 28 and March 1, 2002. Alexandria, VA.

Australian Museum Fish Site. 2003.
http://www.austmus.gov.au/fishes/
Accessed by David Bergendorf (USFWS) January 29, 2004.

Barbier, P., Guise, S., Huitorel, P., Amade, P., Pesando, D., Briand, C. and Peyrot, V. 2001. Caulerpenyne from *Caulerpa taxifolia* has an antiproliferative activity on tumor cell line SK-N-SH and modifies the microtubule network. Life Sciences 70: 415-429.

Bellan-Santini, D., Arnaud, P.M., Bellan, G. and Verlaque, M. 1996. The influence of the introduced tropical alga *Caulerpa taxifolia*, on the biodiversity of the Mediterranean marine biota. Journal of the Marine Biology Association, U.K. 76: 235-237.

California State Water Resources Control Board (CSWRCB). 2003. "*Caulerpa taxifolia*":
http://www.swrcb.ca.gov/rwqcb9/programs/caulerpa/caulerpa.html
Accessed by David Bergendorf (USFWS) January 15, 2004.

Carruthers, T.J.B., Walker, D.I. and Huisman, J.M. 1993. Culture studies on two morphological types of *Caulerpa* (Chlorophyta) from Perth, Western Australia, with a description of a new species. Botanica Marina 36: 589-596.

Ceccherelli, G. and Piazzi, L. 2001. Dispersal of *Caulerpa racemosa* Fragments in the Mediterranean: Lack of detachment time effect on establishment. Botanica Marina 44: 209-213.

Ceccherelli, Giulia. 2002. The spread of *Caulerpa taxifolia* in the Mediterranean: Dispersal strategy, interactions with native species, and competitive ability. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Chen, James C.W. and Jacobs, W.P. 1966. Quantitative study of development of the giant coenocyte, *Caulerpa prolifera*. American Journal of Botany 53(5): 413-423.
Chimino, Guido and Ghiselin, T. 1998. Chemical defense and evolution in the Sacoglossa (Mollusca: Gastropoda: Opisthobranchia). Chemoecology 8: 51-60.

Chisholm, J.R.M., Dauga, C., Ageron, E., Grimont, P.A.D. and Jaubert, J.M. 1996. 'Roots' in mixotrophic algae. Nature 381: 382.

Chisholm, J.R.M. and Jaubert, J.M. 1997. Photoautotrophic metabolism of *Caulerpa taxifolia* (Chlorophyta) in the NW Mediterranean. Marine Ecology Progress Series 153: 113-123.

Chisholm, John R.M., Marchioretti, Manuel and Jaubert, Jean M. 2000. Effect of low water temperature on metabolism and growth of a subtropical strain of *Caulerpa taxifolia* (Chlorophyta). Marine Ecology Progress Series 201: 189-198.

Chisholm, John R. M. and Moulin, Pascale. 2003. Stimulation of nitrogen fixation in refractory organic sediments by *Caulerpa taxifolia* (Chlorophyta). Limnology and Oceanography 48(2): 787-794.

Collado-Vides, L. 2002. Clonal architecture in macroalgae: ecological and evolutionary persepectives. Evolutionary Ecology 15: 531-545.

Collado-Vides, Liga and Robledo, Daniel. 1999. Morphology and photosynthesis of *Caulerpa* (Chlorophyta) in relation to growth form. Journal of Phycology 35: 325-330.

Collado-Vides, L. and Ruesink, Jennifer. 2002. Morphological plasticity and invasive potential of some *Caulerpa* species. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Davis, A.R., Roberts, D.E. and Cummins, S.P. 1997. Rapid invasion of a sponge-dominated deep-reef by *Caulerpa scalpelliformis* (Chlorophyta) in Botany Bay, New South Wales. Australian Journal of Ecology 22: 146-150.

De Haro, Luc., Jouglard, Jacqueline, Thomas, Michele and David, Jean-Marc. 1994. First international workshop on *Caulerpa taxifolia*. Boudouresque C.F, Meinesz, A. & Gravez, V (eds), GIS Posidonie publ.

De Lara-Isassi, Graciella, Alvarez-Hernandez, Sergio and Collado-Vides, Ligia. 2000. Ichtyotoxic activity of extracts from Mexican marine macroalgae. Journal of Applied Phycology 12: 45-52.

Dumay, O., Pergent, G., Pergent-Martini, C. and Amade, P. 2002a. Variations in Caulerpenyne contents in *Caulerpa taxifolia* and *Caulerpa racemosa*. Journal of Chemical Ecology, 28 (2) 343-352.

Dumay, Oliver, Fernandez, Catherine and Pergent, Gerard. 2002b. Primary production and vegetative cycle in *Posidonia oceanica* when in competition with the green algae *Caulerpa taxifolia* and *Caulerpa racemosa*. Journal of Marine Biological Association of the United Kingdom 82: 379-387.

European Environment Agency (EEA). 2004. Fishing fleet trends, 2002. http://themes.eea.eu.int/Sectors_and_activities/fishery/indicators/fishery_fleet Accessed by David Bergendorf (USFWS) August 23, 2004.

Fama, P., Wysor, B., Kooistra, W.H.C.F. and Zuccarello, G.C. 2002a. Molecular Phylogeny of the genus *Caulerpa* (Caulerpales, Cholorphyta) inferred from the chloroplast *tuf*A gene. Journal of *Phycology* 38: 1040-1050.

Fama, Patrizia, Olsen, Jeanine L., Stam, Wytze, T. and Procaccini, Gabriele. 2000b. High levels of intra- and inter-individual polymorphism in the rDNA ITS1 of *Caulerpa racemosa* (Chlorophyta). European Journal of Phycology 35: 349-356.

Ferrer, Ester, Gareta, Amelia Gomez and Ribera, M. Antonia. 1997. Effect of *Caulerpa taxifolia* on the productivity of two Mediterranean macrophytes. Marine Ecology Progress Series 149: 279-287.

Florida Department of Environmental Protection (FDEP). 2003. *Caulerpa spp.* http://www.dep.state.fl.us/southeast/hottopics/caulerpa/caulerpamain.htm Accessed by David Bergendorf (USFWS) January 28, 2004.

Francour, P., Harmelin-Vivien, M., Harmelin, J.G. and J. Duclerc. 1995. Impact of *Caulerpa taxifolia* colonization on the littoral ichthyofauna of North-Western Mediterranean sea: preliminary results. Hydrobiologica 300/301: 345-353.

Frish, Susan M. 2003. Taxonomic Diversity, Geographic Distribution, and Commercial Availability of Aquarium-Traded Species of *Caulerpa* (Chlorophyta, Caulerpaceae) in Southern California, USA. Thesis presented to California State University, Fullerton.

Frish, Susan M. and Murray, Steven N. 2002. The availability of species of *Caulerpa* and "live rock" in retail aquarium outlets in southern California. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Fujita, Y., Itoh, S., Mimour, M., Aizawa, K., Ikeya, T., Kim, S-J. and Murakami, A. 2003. Division of Bioenergetics Annual Report 1994.

Gentner, Brad, Price, Michael and Steinback, Scott. 2001a. Marine Angler Expenditures in the Sotheast Region, 1999. NOAA Technical Memorandum NMFS-F/SPO-48. http://www.rbff.org/research/SEMarine.pdf

Gentner, Brad, Price, Michael and Steinback, Scott. 2001b. Marine Angler Expenditures in the Pacific Coast Region, 2000. NOAA Technical Memorandum NMFS-F/SPO-49. http://www.st.nmfs.gov/st1/econ/NMFS_F_SPO_49rev.pdf

Gravez, Vincent. 2003. The expansion of two marine algas [algae] of the *Caulerpa* kind in the Mediterranean, California and Australia. Translated by MS Internet Explorer. http://www.aquaticecology.org/article.php3?id_article=18 Accessed by David Bergendorf (USFWS) January 8, 2004.

Guiry, M.D. & Nic Dhonncha, E., 2004. AlgaeBase version 2.1. World-wide electronic publication, National University of Ireland, Galway. http://www.algaebase.org Accessed by David Bergendorf (USFWS) December, 2003 - September, 2004.

Harbor Branch Oceanographic Institute (HBOI). 2000. "Killer Alga" Could be Endangering Reefs. Press release on May 23, 2000.

Harbor Branch Oceanographic Institute (HBOI), Ft. Pierce, FL. 2003. Coral Smothering "Green Tide" Seaweed Spreading on Florida Reefs. Press release on June 19, 2003.

IPTEK, Cakrawala. 2004. Species of Algae. http://www.iptek.net.id/eng/cakra_alga/alga_idx.php Accessed by David Bergendorf (USFWS) on July 1, 2004.

Jaubert, Jean M., Chisholm, John R.M., Ducrot, Danielle, Ripley, Herb T., Roy, Laura and Passero-Seitre, Gilles. 1999. No deleterious alterations in *Posidonia* beds in the Bay of Menton (France) eight years after *Caulerpa taxifolia* colonization. Journal of Phycology 35: 1113-1119.

Johnston, C.S. 1969. Studies on the ecological and primary production of Canary Islands marine algae. Proceedings of the International Seaweed Symposium 6: 213-222.

Jousson, O., Pawlowski, J., Zaninetti, L., Zechman, F.W., Dini, F., Di Guiseppe, G., Woodfield, R., Millar, A. and Meinesz, A. 2000. Invasive algae reaches California. Nature 408: 157.

Jung, V., Thibaut, T., Meinesz, A., Phonert, G. 2002. Comparision of the wound-activated transformation of caulerpenyne by invasive and noninvasive *Caulerpa* species of the Mediterranean. Abstract. Journal of Chemical Ecology 28: 2 2091-2105.

Kajimura, Mitsuo. 1977. On dioecious and isogamous reproduction of *Caulerpa scalpelliformis* (R. Br.) Ag. var. *denticulata* (Decsn.) Weber Van Bosse from the Oki Islands, Shimane Prefecture. Bulletion of the Japanese Society of Phycology 25: 27-33.

Khaleafa, A.F., Mohsen, A.F. and Shaalan, S.H. 1982. Effect of different light intensities on growth, amino-acid, fat and sugar concentrations in *Caulerpa prolifera* (Foerskal) Lamouroux. Hydrobiological Bulletin 16 (2-3): 207-212.

Kohler, E. 2003. "Re: Lobiger serradifalci from the Mediterranean". Sea Slug Forum. (November 11, 2001): http://seaslugfroum.net/display.cfm?base=lobiserr&id=5659 Accessed by David Bergendorf (USFWS) December 17, 2003.

Komatsu, Teruhisa, Ishikawa, Tomoji, Yamaguchi, Nobuyuki, Hori, Yukiko and Ohba, Hideo. 2003. But next time?: unsuccessful establishment of the Mediterranean strain of the green seaweed *Caulerpa taxifolia* in the Sea of Japan. Biological Invasions 5: 275-277.

Komatsu, T., Meinesz, A., Buckles, D. 1997. Temperature and light responses of alga *Caulerpa taxifolia* introduced in the Mediterranean Sea. Abstract. Marine Ecology Progress Series 146: 145-153.

Lapointe, Brian E. and Barile, Peter. 2001. Discrimination of nitrogen sources to harmful macroalgal blooms on coral reefs off southeast Florida. Final Report to Florida Institute of Oceanography, subagreement # 4710173L3A.

Lapointe, Brian E. and Yentsch, Charles S. 2003. Progress Report: Physiology and Ecology of Macroalgal Blooms on Coral Reefs off Southeast Florida. EPA Grant Number: R830414.

Lemee, R., Pesando, D., Durand-Clement, M., Dubreuil, A., Meinesz, A., Gurriero, A. and Pietra, F. 1993. Preliminary survey of toxicity of the green alga *Caulerpa taxifolia* introduced into the Mediterranean. Journal of applied Phycology 5: 485-493.

MADL, Pierre and YIP, Maricela. 2003. Literature Review on the Aquarium Strain of *Caulerpa taxifolia*. http://www.sbg.ac.at/ipk/avstudio/pierofun/ct/ct-1.htm
Accessed by David Bergendorf (USFWS) December 15, 2003.

Makowa, J. 2000. Monterey Bay National Marine Sanctuary Fact Sheet: *Caulerpa taxifolia*. July 10, 2003. http://bonita.mbnms.nos.noaa.gov/Research/techreports/Caulerpa.html
Accessed by David Bergendorf (USFWS) December 15, 2003.

Mediterranean Action Plan (MAP). 1998. Report on the workshop on invasive *Caulerpa* species in the Mediterranean. Heraklion, Crete, Greece, March 18-20, 1998.

Meinesz, A. 1999. Killer Algae: The True Tale of a Biological Invasion. University of Chicago Press, Chicago.

Meinesz, Alexandre. 2002a. Introduction for the international *Caulerpa taxifolia* conference. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Meinesz, Alexandre. 2002b. Summary of *Caulerpa taxifolia* invasion and management in the [Mediterranean]. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Merriam-Webster. 2004. Online Dictionary; Merriam-Webster, Inc.
http://www.m-w.com/netdict.htm
Accessed by David Bergendorf (USFWS) August 16, 2004.

Meusnier, I., Olsen, J.L., Stam, W.T., Destombe, C. and Valero, M. 2001. Phylogenetic analyses of *Caulerpa taxifolia* (Chlorophyta) and of its associated bacterial microflora provide clues to the origin of the Mediterranean introduction. Molecular Ecology 10: 931-946.

Millar, Alan. 2002. The introduction of *Caulerpa taxifolia* in New South Wales, Australia. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

National Marine Fisheries Service (NMFS). 2004. Personal communication from the National Marine Fisheries Service, Fisheries Statistics and Economics Division, Silver Spring, MD http://www.st.nmfs.gov/st1/commercial/landings/annual_landings.html
Accessed by David Bergendorf (USFWS) August 23, 2004.

National Oceanic and Atmospheric Administration (NOAA). 2004. National Oceanic Data Center; Coastal Water Temperature Guide.
http://www.nodc.noaa.gov/dsdt/cwtg/index.html
Accessed by David Bergendorf (USFWS) December 23, 2004.

New South Wales Department of Primary Industries; Fisheries (NSWF). 2004. *Caulerpa taxifolia*. http://www.fisheries.nsw.gov.au/thr/species/fn-caulerpa-fishnote.htm
Accessed by David Bergendorf (USFWS) August 31, 2004.

Oklahoma Department of Wildlife Conservation (ODWC). 2004. Oklahoma Administrative Code 800; Subchapter 3. Noxious Aquatic Plants.
http://www.wildlifedepartment.com/aquaticplants.htm

Olsen, Jeanie L., Valero, Myriam, Meusnier, Isabelle, Boele-Bos, Stella and Stam, Wytze T. 1998. Mediterranean *Caulerpa taxifolia* and *C. mexicana* (Chlorophyta) are not conspecific. Journal of Phycology 34: 850-856.

Paul, Valerie J. 2002. Natural products chemistry and chemical ecology of *Caulerpa* spp. with an emphasis on invasive *Caulerpa taxifolia*. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Paul, Valerie J. and Fenical, William. 1986. Chemical defense in tropical green algae, order Caulerpales. Marine Ecology Progress Series 34: 157-169.

Paul, Valerie and Hay, Mark E. 1986. Seaweed susceptibility to herbivory: chemical and morphological correlates. Marine Ecology Progress Series 33: 255-264.

Phillips, J., Price, I. 2002. How different is Mediterranean *Caulerpa taxifolia* (Caulerpales: Chlorophyta) to other populations of the species? Marine Ecology Progress Series 283: 61-71.

Piazzi, Luigi, Balata, David, Ceccherelli, Guilia and Cinelli, Francesco. 2001a. Comparative study of the growth of the two co-occurring introduced green algae *Caulerpa taxifolia* and *Caulerpa racemosa* along the Tuscan coast (Italy, western Mediterranean). Cryptogamie, algologie 22(4): 459-466.

Piazzi, Luigi, Ceccherelli, Giulia and Cinelli, Francesco. 2001b. Threat to macroalgal diversity: effects of the introduced green alga *Caulerpa racemosa* in the Mediterranean. Marine Ecology Progress Series 210: 149-159.

Piazzi, Luigi, Balata, David, Cecchi, Enrico and Cinelli, Francesco. 2003. Co-occurrence of *Caulerpa taxifolia* and *C. racemosa* in the Mediterranean Sea: interspecific interactions and influence on native macroalgal assemblages. Cryptogamie Algologie 24(3): 233-243.

Raloff, J. (July 4, 1998). "Rouge Algae. The Mediterranean floor is being carpeted with a shaggy, aggressive invader." ScienceNewsOnline. July 21, 2003. www.sciencenews.org/sn_arc98/7_4_98/bob1.htm
Accessed by David Bergendorf (USFWS) December 23, 2004.

Renoncourt, Line and Meinesa, Alexandre. 2002. Formation of propagules on an invasive strain of *Caulerpa racemosa* (Chlorophyta) in the Mediterranean Sea. Phycologia 41(5): 533-535.

Riechert, R. and Dawes, C.J. 1986. Acclimation of the green alga *Caulerpa racemosa* var. *uvifera* to light. Botanica Marina 29: 533-537.

Silva, Paul C. 2002. An overview of the genus *Caulerpa*. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Silva, Paul C. 2003. Historical overview of the genus *Caulerpa*. Cryptogamie Algologie 24(1): 33-50.

Silva, Paul C. (research botanist, University of California at Berkeley). 2004. Personal communication to David Bergendorf, USFWS, Stockton California on January 16, 2004.

Smith, Celia M. and Walters, Linda J. 1999. Fragmentation as a strategy for *Caulerpa* species: Fates of fragments and implications for management of an invasive weed. P.S.Z.N. Marine Ecology 20(2-4): 307-319.

South Carolina Department of Natural Resources (SCDNR). 2004. Illegal Aquatic Plants in South Carolina. http://www.dnr.state.sc.us/water/envaff/aquatic/illegal1.html
Accessed by David Bergendorf (USFWS) August 13, 2004.

Southern California *Caulerpa* Action Team (SCCAT). 2003a. Southern California *Caulerpa* Action Team, *Caulerpa* Database. www.sccat.net
Accessed by David Bergendorf (USFWS) December 17, 2004.

Southern California *Caulerpa* Action Team (SCCAT). 2003b.
Status Report: *Caulerpa taxifolia* Invasions in Southern California. www.sccat.net
Accessed by David Bergendorf (USFWS) December 17, 2004.

Southern California Wetlands Recovery Project (SCWRP). 2003. Information item: Invasion of *Caulerpa taxifolia* to Southern California. Wetlands Managers Group Report 2000. http://www.coastalconservancy.ca.gov/scwrp/BOGMtgs/BOG102000/Caulerpa.htm Accessed by David Bergendorf (USFWS) January 8, 2004.

Stazione Zoologica Anton Dohrn. 1997. Biodiversity, Dynamics and Interaction in Benthic System: Seagrass Ecosystem. Activity Report 1996/1997. http://www.szn.it/actrep/aqua_206.htm Accessed by David Bergendorf (USFWS) December 22, 2004.

Terrados, J. 1991. Crecimiento y produccion de las praderas de macrofitos del Mar Menor. Ph. D. Thesis. University of Murcia. [As cited in: Terrados, Jorge and Ros, Joandomenec. 1991. Production dynamics in a macrophyte-dominated ecosystem: The Mar Menor coastal lagoon (SE Spain). Oecologia aquatica 10: 255-270.]

Terrados, Jorge and Ros, Joandomenec. 1991. Production dynamics in a macrophyte-dominated ecosystem: The Mar Menor coastal lagoon (SE Spain). Oecologia aquatica 10: 255-270.

Terrados, J. and Ros, J.D. 1992. The influence of temperature on seasonal variation of *Caulerpa prolifera* (Forsskal) Lamouroux photosynthesis and respiration. Journal of Experimental Marine Biological Ecology 162: 199-212.

Thibaut, Thierry and Meinesz, Alexandre. 2000. Are the Mediterranean ascoglossan molluscs *Oxynoe olivacea* and *Lobiger serradifalci* suitable agents for a biological control against the invading tropical alga *Caulerpa taxifolia*? Life Sciences 323: 477-488.

Thibaut, Thierry and Meinesz, Alexandre. 2002. Discussion of *Caulerpa taxifolia* management in the Mediterranean. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Thomas, Jane. 2003. *Caulerpa taxifolia* in Moreton Bay- Distribution and Seagrass Interactions. Thesis submitted to the Department of Botany, The University of Queensland, Australia.

Tippets, Karoly. 2002. The effects of *Caulerpa taxifolia* on invertebrate abundance in Agua Hedionda Lagoon, California. IN: Latto, John, Orr, Matt, Remais, Justin and Desai, Manish (eds.) 2002. The Environment: Studies of Ecological Interconnections. University of California at Berkeley. http://istsocrates.berkeley.edu/~es196/projects/2002final/introduction.htm Accessed by David Bergendorf (USFWS) August 31, 2004.

Turkish Daily News (TDN). March 17, 1998. Turkey to attend 'terrorist moss' summit. http://www.turkishdailynews.com/old_editions/03_17_98/for.htm

United Nations Food and Agriculture Organization (UN FAO). 2004. Fisheries Global Information System: Fisheries Statistics. http://www.fao.org/figis/servlet/static?dom=root&xml=tseries/index.xml Accessed by David Bergendorf (USFWS) August 23, 2004.

United Nations Food and Agriculture Organization (UN FAO). 2002. The State of World Fisheries and Aquaculture. http://www.fao.org/docrep/005/y7300e/y7300e00.htm Accessed by David Bergendorf (USFWS) August 24, 2004.

U.S. Department of the Interior, Fish and Wildlife Service, U.S. Department of Commerce and U.S. Census Bureau (USDOI). 2001. National Survey of Fishing, Hunting and Wildlife-Associated Recreation. http://www.census.gov/prod/2002pubs/fhw01-ca.pdf

University of Hawai'i at Manoa (UHM). 2004. Botany. http://www.botany.hawaii.edu/ReefAlgae/caulerpa.htm Accessed by David Bergendorf (USFWS) February 12, 2004.

Vest, S.E., Dawes, C.J. and Romeo, J.T. 1983. Distribution of caulerpin and caulerpicin in eight species of the green alga *Caulerpa* (Caulerpales). Botanica Marina 26: 313-316.

Vroom, Peter S. and Smith, Celia M. 2001. The Challenge of Siphonous Green Algae. American Scientist 89: 525-531.

Williams, Susan L. 1984a. Decomposition of the tropical macroalga *Caulerpa cupressoides* (West) C. Agardh: Field and laboratory studies. Journal of Experimental Marine Biological Ecology 80: 109-124.

Williams, Susan Lynn. 1984b. Uptake of sediment ammonium and translocation in a marine green macroalga *Caulerpa cupressoides*. Limnology and Oceanography 29(2): 374-379.

Williams, S.L., V.A. Breda, T.W. Anderson, B.B. Nyden. 1985. Growth and sediment disturbances of *Caulerpa* spp. (Chlorophyta) in a submarine canyon. Marine Ecology Progress Series 21:275-281.

Williams, S.L. and W.C. Dennison. 1990. Light availability and diurnal growth of a green macroalga (*Caulerpa cupressoides*) and a seagrass (*Halophila decipiens*). Mar. Biol. 106:437-443.

Williams, S., Grosholz, E. 2002. Preliminary reports from the *Caulerpa taxifolia* invasion in southern California. Marine Ecology Progress Series 233, 307-310.

Woodfield, R. 2003. Noxious seaweed found in southern California coastal waters. Southwest Regional Office. National Marine Fisheries Service. http://swr.nmfs.noaa.gov/hcd/caulerpa.htm
Accessed by David Bergendorf (USFWS) December 29, 2003.

Woodfield, Rachel and Merkel, Keith. 2004. Eradication and Surveillance of *Caulerpa taxifolia* within Agua Hedionda Lagoon, Carlsbad, California: Third Year Status Report.http://swr.nmfs.noaa.gov/hcd/caulerpa/AguaHediondaYear%203%20Status%20Report.pdf
Accessed by David Bergendorf (USFWS) August 30, 2004.

Yip, M. 2001. "Essay about *Caulerpa taxifolia*." Marine Biology I. July 10, 2003. http://sbg.ac.at/ipk/avstudio/pierofun/ct/caulerpa.htm
Accessed by David Bergendorf (USFWS) December 29, 2003.

Zenetos, Argyro, Todorova, Valentina, and Alexandrov, Boris. 2003. Marine biodiversity changes in the Mediterranean and Black Sea regions. Extended Abstract: International Conference on the Sustainable Development of the Mediterranean and Black Sea Environment.
http://www.iasonnet.gr/abstracts/zenetos.html
Accessed by David Bergendorf (USFWS) August 23, 2004.

Zuljevic, Ante and Antolic, Boris. 2002a. Appearance and eradication of *Caulerpa taxifolia* in Croatia. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Zuljevic, Ante and Antolic, Boris. 2002b. Reproduction of *Caulerpa taxifolia* in the Mediterranean Sea. IN: International *Caulerpa taxifolia* Conference Proceedings January 31-February 1, 2002. San Diego, CA, U.S.A.

Zuljevic, A., Thibaut, T., Elloukal, H., Meinesz, A. 2001. Sea slug disperses the invasive *Caulerpa taxifolia*. Short Communications. Journal of the Marine Biological Association of the United Kingdom 81.

Personal Communications:

Lapointe, Brian E. (Harbor Branch Oceanographic Institution). January 29, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Millar, Alan (Principal Research Scientist; Royal Botanic Gardens, Sydney, Australia). July 5, 2004 and August 30, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Peyton, Kim (University of Hawaii). September 28, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Silva, Paul (Research Botanist; University of California at Berkeley). January 18, 2004 and August 31, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Walters, Linda (Professor of Biology, University of Central Florida). August 25 and September 27, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Williams, Susan (Director of Bodega Marine Laboratory). August 19, 2004. Personal communication to David Bergendorf, USFWS, Stockton, CA.

Woodfield, Rachel (Senior Biologist, Merkel & Associates, Inc.). Personal communication to David Bergendorf (U.S. Fish and Wildlife Service) on January 28, 2004.

Picture Credits:

Figure 1. *Caulerpa taxifolia* exhibits a habit typical of the genus: a creeping rhizome produces rhizoids downward and photosynthetic branches (in this case bearing leaf-like pinnules) upward (Drawing by Alan Millar, Royal Botanic Garden, Sydney, Australia).

Figure 2. On the Pacific coast of North America indigenous populations of *Caulerpa* only exist as far north as Guadalupe Island (Map from San Diego Natural History Museum).

Figure 3. Caulerpa taxifolia, prior to treatment in California (Photo by R. Woodfield, Merkel and Associates 2000).

Figure 4. *Caulerpa brachypus* growing on coral in FL (Photo by FDEP 2003).

Figure 5. A rapidly growing form of *Caulerpa racemosa* (Photo by R. Woodfield, Merkel and Assoc.)

Figure 6. *Caulerpa prolifera* (photo by G. Felicini: IN Guiry and Dhonncha 2004).

Figure 7. *Caulerpa cupressoides* (Photo by Iris Bonig: IN Guiry and Dhonncha 2004).

Figure 8. *Caulerpa scalpelliformis* (Photo by John Huisman: IN Guiry and Dhonncha 2004).

Figure 9. *Caulerpa mexicana* (Photo by John Huisman: IN Guiry and Dhonncha 2004).

Figure 10. *Caulerpa verticillata* (Photo by John Huisman: IN Guiry and Dhonncha 2004).

Figure 11. *Caulerpa sertularioides* (Photo by John Huisman: IN Guiry and Dhonncha 2004).

Appendix A. A Prevention Program for the Mediterranean Strain of *Caulerpa taxifolia*

Submitted to the
Aquatic Nuisance Species Task Force

By the

Caulerpa taxifolia Prevention committee

Prepared by

Sandra M. Keppner
U.S. Fish and Wildlife Service
Lower Great Lakes Fishery Resources Office
405 North French Road, Suite 120A
Amherst, NY 14228

February 11, 2002

Executive Summary

In October 1998, the Department of Interior was alerted to potential threats posed an invasive, aquarium-bred, tropical green seaweed known as *Caulerpa taxifolia*. The U.S. Fish and Wildlife Service (Service) was assigned leadership and asked to conduct an investigation of this potential invader. The Service completed its investigation and presented findings to the national Aquatic Nuisance Species (ANS) Task Force in November 1998.

Since its discovery off the coast of Monaco in the Mediterranean in 1984, *C. taxifolia* has spread rapidly severely impacting the coastal ecosystems of six Mediterranean countries (Croatia, France, Italy, Monaco, Spain, and Tunisia). It is believed to have originated from an aquarium, as it is genetically similar to strains cultured and propagated in western European aquaria since the early 1970's. The introduced *C. taxifolia* exhibits characteristics and tolerances different from those exhibited by this species in its native habitat. For example, the Mediterranean strain demonstrates only asexual reproductive strategies, forms dense mats rather than growing in small isolated clumps, reaches much greater heights in terms of growth, and tolerates a wider range of temperatures. Based on thermal tolerances, its potential distribution in the U.S. is predicted to include coastal areas south of Virginia Beach, Virginia on the Atlantic coast, areas south of Stonewall Bank, Oregon on the Pacific coast, the Gulf of Mexico, Hawaii, Puerto Rico, the U.S. Virgin Islands, American Somoa, Guam, and the Northern Mariana Islands. The potential expansion of *C. taxifolia* throughout these waters threatens native flora and fauna, especially threatened and endangered species, and may pose potential economic impacts on revenues generated by commercial and recreational fisheries, tourism, and industrial facilities.

In November 1998, the ANS Task Force determined that the risk of an unintentional introduction of *C. taxifolia* (Mediterranean strain) through identified pathways was significant and the adverse consequences of an introduction would likely be substantial. According to provisions of the Nonindigenous Aquatic Nuisance Prevention and Control Act of 1990, these findings warrant the development and implementation of a cooperative comprehensive prevention program to minimize the risk of introduction. As requested by the ANS Task Force, the Service and the U.S. Department of Agriculture's Animal and Plant Health Inspection Service drafted *A Prevention Program for the Mediterranean Strain of Caulerpa taxifolia*. The Prevention Program was presented to the ANS Task Force in August 1999. The ANS Task Force established a *Caulerpa taxifolia* Prevention Committee to review and refine the Prevention Program and oversee implementation.

This revised Prevention Program reflects the opinions of Committee members as well as the subsequent changes in priorities following the discovery of *Caulerpa taxifolia* in a California coastal lagoon north of San Diego, CA. As mandated by the Nonindigenous Aquatic Nuisance Prevention and Control Act, the Prevention Program is comprehensive, consisting of eight objectives and subsequent recommendations. The overall goal of the Prevention Program is to prevent the introduction, establishment, and dispersal of the invasive Mediterranean strain of *Caulerpa taxifolia* in U.S. waters. The objectives address Coordination and Leadership; Dispersal Mechanisms and Pathways Analysis; Surveillance and Detection; Control; Research;

Regulation; Legislation; and Education. The recommended actions include both short- and long-term initiatives that will require cooperation between international, federal, state, tribal, private and public organizations. It is also recognized that this Prevention Program is limited by current knowledge and information gaps. This Prevention Program will be updated as necessary as new information becomes available.

It is the hope of the Committee that through cooperation, coordination, and commitment, this Prevention Program will be implemented to effectively protect U.S. coastal resources from further invasions of *C. taxifolia*. Furthermore, the Committee hopes that the knowledge, awareness, and experience gained through implementation will serve as a model leading to advanced prevention planning for other invasive species, including other species of *Caulerpa* that may threaten coastal resources.

Table of Contents

A Prevention Program for the Mediterranean Strain of *Caulerpa taxifolia*

INTRODUCTION

In October 1998, the Department of Interior was alerted to the invasiveness of an aquarium-bred clone of tropical green seaweed known as *Caulerpa taxifolia* and that this species posed a potential threat to U.S. coastal waters. In a letter signed by over 100 researchers, the scientific community requested that the Secretary of Interior (Secretary) initiate immediate action to prevent this strain of *C. taxifolia* from entering and becoming established in U.S. waters. This species had already severely impacted the coastal ecosystems of five Mediterranean countries (Croatia, France, Italy, Monaco, and Spain), forming dense mats, and eliminating biodiversity. The European invasion was well publicized, as controversy surrounded its origin and response by authorities. This publicity provoked a response in the U.S., and fortunately, an awareness among scientists and resource agencies of the risk of complacency. The Secretary assigned leadership to the U.S. Fish and Wildlife Service (Service) and requested an investigation of *C. taxifolia*[1] and its potential for establishment in U.S. waters.

In November 1998, the Service presented its findings in the report, *Caulerpa taxifolia: A Potential Threat to U.S. Coastal Waters* to the Aquatic Nuisance Species (ANS) Task Force (Keppner et al. 1998) (Appendix A). The ANS Task Force, an intergovernmental body established by the Nonindigenous Aquatic Nuisance Prevention and Control Act of 1990, provided an appropriate forum for further discussion of *C. taxifolia,* including its potential impacts on U.S. coastal ecosystems and the need for prevention. Based on the report findings, the ANS Task Force determined that the risk of an unintentional introduction of *C. taxifolia* through identified pathways was significant and that the adverse consequences of an introduction would likely be substantial. According to provisions of the Nonindigenous Aquatic Nuisance Prevention and Control Act of 1990, these findings warranted the development and implementation of a cooperative comprehensive prevention program to minimize the risk of introduction (Appendix B).

The ANS Task Force requested that the Service and the U.S. Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS) jointly develop a comprehensive program to prevent the introduction and establishment of *C. taxifolia* in U.S. waters. These agencies subsequently drafted *A Prevention Program for the Mediterranean Strain of Caulerpa taxifolia* (Prevention Program) and presented it to the ANS Task Force in August, 1999 (Keppner and Caplen 1999). The draft prevention Program consisted of nine components. Actions were recommended within each

[1] For the purposes of this report, *C. taxifolia* refers to the invasive Mediterranean strain unless otherwise noted.

component to achieve the overall goal of preventing the introduction, establishment, and dispersal of the invasive strain of *C. taxifolia* in U.S. waters. The ANS Task Force established a *Caulerpa taxifolia* Prevention Committee (Committee) to review and refine the Prevention Program, and oversee implementation. Representatives from various affected entities, including state and federal resource agencies, research, education, and industries such as large aquaria and zoos, pet and small aquaria, shipping, seafood, and bait were invited to participate on the Committee (Appendix C). Committee members met for the first time in April 2000 to review the draft Prevention Program and prioritize recommended actions. To maintain comprehensiveness and flexibility, the Committee chose to develop short-term Action Plans as a means of implementing the Prevention Program. The Action Plans will identify urgent or immediate needs and priorities, outline specific actions, and consider current research findings, funding needs, and agency commitments.

The Committee's decision to ensure the Prevention Program would be comprehensive and dynamic was substantiated just two months later when *C. taxifolia* was discovered off the coast of California (Anderson and Keppner 2001a; 2001b). The Committee responded providing guidance, assistance, and national leadership to local and regional governments and organizations planning eradication efforts. The first Action Plan was drafted with a new perspective on prioritization. Development of the Action Plan was augmented by the workshop, *Caulerpa taxifolia: Implementing a National Prevention Program*, sponsored by the Service and the Committee. The workshop, held in San Diego, CA in July 2001, brought together scientists, resource agencies, policy-makers, and control experts to discuss priority needs, learning from the California response. In January 2002, the Service, in cooperation with California Sea Grant and the California Department of Fish and Game, sponsored the International *Caulerpa taxifolia* Conference. Scientists and resource managers from Australia, France, Italy, Croatia, New Zealand, South Africa, Mexico, and the U.S. gathered in San Diego, CA for two days to discuss science-based control, management, and eradication of *Caulerpa taxifolia*.

This revised Prevention Program reflects the opinions of its members as expressed at the April 2000 meeting as well as newly identified needs raised at the 2001 workshop and the 2002 conference. As mandated by the Nonindigenous Aquatic Nuisance Prevention and Control Act, the Prevention Program is comprehensive, however, the objectives and actions outlined have been reviewed in terms of practical and realistic means. It includes both short- and long-term actions, many that will require commitment and partnership among multiple agencies and organizations. It is the hope of the Committee that through cooperation, coordination, and commitment, this Prevention Program will be implemented to effectively protect U.S. coastal resources from further invasions of *C. taxifolia*.

BACKGROUND

The tropical green alga *C. taxifolia* was first detected in the Mediterranean Sea in 1984 (Meinesz 1999; Meinesz et al. 1993). According to genetic findings, this invasive strain is believed to have originated from *C. taxifolia* plants bred in western European aquaria (Jousson et al. 1998). Furthermore, recent genetic analyses indicate that the source population of the aquarium plants in Europe is Brisbane, Australia, and that the population in Brisbane is likely the result of an introduction from Northern Australia (Olsen et al. 2002). The introduced *C. taxifolia* exhibits characteristics and tolerances different from those exhibited in its native habitat. For example, the Mediterranean strain demonstrates only asexual reproductive strategies, forms dense mats rather than growing in small isolated clumps, reaches much greater heights in terms of growth, and tolerates a wider range of temperatures, surviving between the isotherms of 10-31ºC (Komatsu et al. 1997; Meinesz et al. 1993; Raloff 1998).

Like many successful invasion species, the Mediterranean strain of *C. taxifolia* spreads rapidly, possessing physiological, morphological, and ecological characteristics that contribute to its ability to spread, displace native species, and alter existing coastal habitat areas. Perhaps contributing most significantly to its ability to proliferate, the Mediterranean strain of *C. taxifolia* exhibits vegetative (asexual) reproduction. Small cuttings or plants fragments are capable of regenerating new plants. In addition, the Mediterranean strain is able to adapt to a wide range of habitat conditions including substrate, light intensity, and water quality (Ceccherelli and Cinelli 1998; Komatsu et al. 1997). *C. taxifolia* synthesizes toxic or repellent secondary metabolites to defend against herbivory and epiphytism (Ferrer et al. 1997; Guerriero et al. 1992). These toxins affect local flora and fauna through both direct and indirect contact (chemical interaction), affording *C. taxifolia* a competitive advantage over native species (Ferrer et al. 1997; Giannotti et al. 1994; Merino et al. 1994).

The invasion of *C. taxifolia* has altered both the flora and fauna endemic to the coastal waters of the Mediterranean, reducing biodiversity. Competition and habitat modification are the two primary mechanisms used by *C. taxifolia* to influence and often exclude local populations. Through competitive interactions (light and space), indigenous flora are reduced or eliminated leading to monocultural stands of *C. taxifolia* (Boudouresque et al. 1992; Ferrer et al. 1997). In addition, the production and release of toxins provides an effective competitive advantage to *C. taxifolia*. The dense mats formed by *C. taxifolia* change the bottom structure of the seafloor affecting benthic fauna (Bellan-Santini et al 1996). Changes in the benthic fauna as well as habitat modifications (e.g. loss of important spawning or nursery areas) likely contribute to observed changes in ichthyofauna (Francour et al. 1995; Harmelin-Vivien et al. 1996).

The potential spread of *C. taxifolia* in U.S. coastal systems is expected to cause similar impacts. Based on thermal tolerances of *C. taxifolia*, its potential distribution in the U.S. is predicted to include coastal areas south of Virginia Beach, Virginia on the Atlantic coast, areas south of Stonewall Bank, Oregon on the Pacific coast, the Gulf of Mexico, Hawaii, Puerto Rico, the U.S. Virgin Islands, American Samoa, Guam, and the Northern Mariana Islands (Keppner et al. 1998). In the absence of natural control mechanisms, invasive species typically expand rapidly, often exponentially. This expansion will threaten native flora and fauna, especially threatened and endangered species. Potential negative economic impacts may include a decrease in revenues generated by commercial and recreational fisheries, tourism, and industrial facilities.

EXISTING AUTHORITIES

FEDERAL

Nonindigenous Aquatic Nuisance Prevention and Control Act (Appendix B)
Enacted in 1990, and reauthorized in 1996 as the National Invasive Species Act (P.L. 104-332. 110 Stat. 4073. 10/26/96), this statute established an intergovernmental mechanism for the development of a national program to: 1) prevent the unintentional introduction and dispersal of nonindigenous species through ballast water management and other requirements; 2) coordinate federally conducted, funded, or authorized research, prevention control, information dissemination and other activities regarding aquatic nuisance species, 3) develop and carry out environmentally sound control methods to prevent, monitor, and control unintentional introductions of nonindigenous species from pathways other than ballast water, 4) understand and minimize economic and ecological impacts of nonindigenous aquatic nuisance species that become established, and 5) establish a program of research and technology development and assistance to states in the management and removal of zebra mussels. The Act established the ANS Task Force, chaired by the Service and the National Oceanic and Atmospheric Administration (NOAA), to coordinate federal efforts and develop the national Aquatic Nuisance Species Program to prevent the introduction and dispersal of aquatic nuisance species, monitor, control, and research such species, and disseminate related information.

The Act directs the ANS Task Force to establish and implement measures within the Aquatic Nuisance Species Program to minimize the risk of introducing aquatic nuisance species into U.S. waters including the identification of pathways by which aquatic organisms are introduced, assessing the risk that an organism may become an aquatic nuisance species, and evaluating prevention strategies. If the ANS Task Force determines that there is a significant risk of an unintentional introduction of an aquatic nuisance species by an identified pathway and that adverse consequences of such an introduction are likely to be substantial, the ANS Task Force shall, acting through the appropriate federal agency, and after opportunity for public comment, carry out cooperative, environmentally sound efforts with regional, state, and local entities to minimize the risk of such an introduction.

Plant Protection Act (Appendix D)
The Plant Protection Act (P.L. 106-224, 7 U.S.C. 7701-7758) authorizes port-of-entry and follow-up activities (for example, quarantine, treatment, disposal, control or eradication programs) by USDA to restrict the introduction and spread of non-native noxious weeds. Under the Act, no person shall import or enter any noxious weed identified in regulation, into or through the United States, or move any noxious weed interstate, unless done in accordance with regulations issued by the Secretary. Any such entry or movement therefore requires a permit from APHIS, and is usually allowed only into an approved and inspected containment facility.

Lacy Act
Originally enacted in 1900, the Lacey Act, as amended in 1981 (P.L. 97-79. 16 U.S.C. 3371-3378) makes it illegal to import, export, sell, receive, acquire, or purchase fish, wildlife, or plants taken, possessed, transported, or sold in violation of U.S. or tribal law. In addition, this Act makes it unlawful to engage in interstate or foreign commerce involving any fish, wildlife, or plant material taken, possessed, transported or sold in violation of state or foreign law. Specific provisions authorize the federal government to prescribe requirements and issue permits for importation of wild animals and birds under humane and healthful conditions. This law may be applicable for regulating the introduction of non-native species, if only indirectly. For example, if a plant whose sale is illegal in one state is purchased in that state and then taken to another state, the purchaser/transporter may have violated the Act, since the federal Lacy Act can be triggered by violations of certain state laws combined with interstate transport. However, if the non-native plant is purchased in a state where its sale is allowed, and then simply brought into another state (e.g., as household effects), it is unclear whether the Lacey Act would apply.

Executive Order 13089 - Coral Reef Protection
President Clinton signed Executive Order 13089 on Coral Reef Protection (63 *Fed. Reg.* 32701, June 16, 1998), on June 11, 1998. This Executive Order seeks to protect coral reef ecosystems in Florida, Hawaii, American Samoa, Guam, the Northern Mariana Islands, Puerto Rico, and the U.S. Virgin Islands through enhanced coordination of federal activities that impact coral reefs. This would include federal initiatives regarding Aquatic Nuisance Species in marine environments.

Executive Order 13112 - Invasive Species
President Clinton signed Executive Order 13112 on Invasive Species (64 *Fed. Reg.* 6183, Feb. 8, 1999), on February 3, 1999, revoking President Carter's 1977 Executive Order 11987 on exotic species. The new Executive Order seeks to prevent the introduction of invasive species and provide for their control and minimize their impacts through better coordination of federal agency efforts under a National Invasive Species Management Plan to be developed by an interagency Invasive Species Council. The order directs all federal agencies to address invasive species concerns as well as refrain from actions likely to increase invasive species problems. The Invasive Species Council, supported by

an advisory committee, is also to develop recommendations for international cooperation, promote a network to document and monitor invasive species impacts, and encourage development of an information-sharing system on invasive species.

In addition, although the following Acts were generally designed to address issues other than invasive species, broad interpretation of these statutes could be applicable to *C. taxifolia* regulation and prevention activities. All applicable provisions should be implemented as appropriate.

- Alien Species Prevention and Enforcement Act of 1992
- Atlantic Coastal Fisheries Cooperative Management Act of 1993, as amended 1996
- Coastal Zone Management Act of 1972, as amended 1975,1976,1978,1986,1990,1992,1996
- Endangered Species Act of 1973, as amended 1976-1982,1984,1988
- Fish and Wildlife Conservation Act of 1980, as amended 1986,1988,1990,1992
- Fish and Wildlife Improvement Act of 1978
- Florida Keys National Marine Sanctuary and Protection Act of 1990
- Interjurisdictional Fisheries Act of 1986, as amended 1990,1992,1993,1994,1996
- Marine Mammal Protection Act of 1972, as amended 1973,1976-1978,1980-1982,1984,1986,1988,1990,1992-1994,1996
- Marine Protection, Research, and Sanctuaries Act: Part I - Marine Sanctuaries of 1974, as amended 1975-1977,1980-1982,1984,1988,1992,1996, 2000
- Marine Resources and Engineering Development Act of 1966, as amended 1966,1968-1970,1986
- National Coastal Monitoring Program of 1992
- National Environmental Education Act of 1990, as amended 1994
- National Environmental Policy Act of 1969, as amended 1975, 1994
- Regional Marine Research Program of 1990

STATE

On September 25, 2001, the State of California enacted legislation (AB No. 1334) prohibiting the sale, possession, import, transport, transfer, release, or giving away of marine algae of the *Caulerpa* species: *taxifolia, cupressoides, mexicana, sertularoides, floridana, ashmeadii, racemosa, verticillata*, and *scapelliformis* (Appendix E). These nine species of *Caulerpa* are either difficult to distinguish from *C. taxifolia* or could become invasive if introduced into marine coastal areas. These regulations provide the state with the authority to confiscate and destroy these species when found. The Department of Fish and Game is authorized to permit possession for research purposes.

LOCAL

The City of San Diego, CA prohibits the sale and possession of all *Caulerpa* species (Chapter 6 Article 3 Division 15 of San Diego Municipal Code) (Appendix F). This represents the most comprehensive ban of *Caulerpa* species, demonstrating the commitment of the city to protect its coastal resources from invasive *Caulerpa* species.

GOAL STATEMENT

Prevent the introduction, establishment, and dispersal of the invasive Mediterranean strain of *Caulerpa taxifolia* in U.S. waters.

OBJECTIVES AND ACTIONS

As mandated by the Nonindigenous Aquatic Nuisance Prevention and Control Act, the Prevention Program is comprehensive, consisting of eight objectives and subsequent recommendations. The objectives address Coordination and Leadership; Dispersal Mechanisms and Pathways Analysis; Surveillance and Detection; Control; Research; Regulation; Legislation; and Education. The recommended actions include both short- and long-term initiatives requiring commitment from and partnership among international, federal, state, tribal, private and public organizations.

To maximize the Committee's flexibility and responsiveness in implementing this Prevention Program, immediate prevention needs and implementation priorities will be identified in short-term Action Plans developed by the Committee. Development of such Action Plans will allow the Committee to consider funding needs, agency responsibilities, geographic and research priorities, the current state of infestation, and new research findings, ensuring implementation of this comprehensive Prevention Program is effective, efficient, and successful.

OBJECTIVE I -- COORDINATION AND LEADERSHIP

Provide national coordination and leadership to federal and state governments and to commercial, private, and public organizations conducting *Caulerpa taxifolia* prevention efforts.

Problem

Implementation of an effective prevention program requires centralized coordination and leadership along with the support and participation of many federal agencies and organizations. Lead agencies will be required to strengthen their working relationships with existing partners and establish new relationships with public and private sector entities that could be impacted by the introduction and/or establishment of *C. taxifolia* in U.S. waters.

Recommended Actions

1. Maintain active membership on the *Caulerpa taxifolia* Prevention Committee of the ANS Task Force to lead and coordinate implementation of the Prevention Program, and refine ongoing prevention activities and the Prevention Program as new scientific information becomes available.

2. Establish Working Groups within the *Caulerpa taxifolia* Prevention Committee, as needed, to address specific issues, objectives, or actions to ensure effective and efficient execution of prevention activities.

OBJECTIVE II -- DISPERSAL MECHANISMS AND PATHWAYS ANALYSIS

Eliminate known and potential pathways of introduction and dispersal contributing to the spread of *Caulerpa taxifolia* in U.S. coastal waters.

Problem

Development of effective prevention strategies for nonindigenous aquatic species like *C. taxifolia* requires a thorough understanding of potential pathways and means of dispersal that could introduce and spread an invasive aquatic species. The most direct and effective prevention strategy is to identify and eliminate known and potential dispersal pathways. Although some dispersal mechanisms are natural and therefore difficult to interrupt, many if not most are related to anthropogenic activities including international, national, regional, and local activities. All of these pathways could potentially facilitate the introduction and spread of *C. taxifolia* into U.S. waters.

C. taxifolia reproduces vegetatively therefore the disruption and subsequent transport of small plant fragments by any means may result in the establishment of a new population. Activities such as dredging, mechanical removal, anchoring, commercial fishing, boating, ocean currents, and storms have all contributed to the expansion of *C. taxifolia* in the Mediterranean.

C. taxifolia is commonly used in the aquarium industry for display purposes. The importation of this species and its eventual release or escape poses a risk to U.S. waters. Other potential pathways for *C. taxifolia* introduction into U.S. waters include use as packing material for imported shipments of fresh seafood, bait imports, and possibly ballast water from transoceanic vessels. Each of these pathways pose a potential threat to U.S. coastal systems if *C. taxifolia* plant fragments survive shipment and are released into U.S. waters. A combination of research and risk analysis is required to determine the pathways that most significantly threaten U.S. waters.

Recommended Actions

1. Conduct a formal pathways analysis and risk assessment to determine the risk of introduction and/or spread posed by various pathways, such as (but not limited to) aquarium escapes/releases, packing material for imported shipments of fresh seafood, bait imports, ballast water from transoceanic vessels, and commercial and recreational fishing and tourism (cruiseline) vessels. Identify high risk geographic and temporal patterns associated with various pathways.

a. Assess current and routine disposal practices for algal packing materials, water, and any other materials contained in shipments.

2. Identify and investigate national and foreign source populations (e.g. aquarium and commercial sources, Internet sales), mechanisms of entry (including postal and other mail services), domestic entry locations, interstate transport mechanisms, and final destinations of imported shipments.

OBJECTIVE III -- SURVEILLANCE AND DETECTION

Coordinate *Caulerpa taxifolia* surveillance efforts to maximize early detection of new populations, track range expansions, and evaluate prevention and control strategies.

Problem

Detection and surveillance are critical components of prevention and control strategies. Management and policy decisions are often based on information gathered through active surveillance programs as the primary source of baseline data on pre-existing populations and habitat. The implementation of surveillance provides managers with an opportunity to: 1) detect newly established populations early, 2) track the expansion or spread of introduced species, 3) predict potential or estimate realized impacts of introductions or range expansions, and 4) evaluate prevention and control strategies.

Like most invasive species, the likelihood of successful control and eradication of *C. taxifolia* is highly dependent on early detection. The various dispersal mechanisms used by *C. taxifolia* require a multi-faceted approach to detection and surveillance. This approach requires cooperation and partnership among federal, state, tribal, commercial, private, and public organizations. The information gained through a cooperative comprehensive approach to detection and surveillance will contribute to refining this Prevention Program.

Recommended Actions

1. Integrate surveillance and detection programs for invasive *C. taxifolia* into ongoing coastal monitoring programs, including permit compliance activities (e.g. U.S. Army Corps of Engineers, National Marine Fisheries Service, Service). Utilize education and outreach strategies to alert scientists, managers, consultants, and the public about the need to watch for this harmful algal species.

a. Prioritize effort in areas where *C. taxifolia* has already been introduced and where dispersal is most likely to occur based on already established invasions or high risk pathways of introduction and spread.

b. Identify researchers and agencies already participating in surveillance or monitoring efforts to advance partnership and cooperation.

c. Designate appropriate federal agency to coordinate surveillance reporting and act as a central clearinghouse for surveillance information. (Report confirmed sitings to the U.S. Geological Survey in Gainesville, Florida and other appropriate algal database managers and invasive species hotlines.) Ensure surveillance information is shared and Internet accessible (Objective VIII, 3).

2. Develop effective and efficient field identification and surveillance protocols to determine the extent of colonization when new populations are discovered and to track expansion. Incorporate surveillance protocols into rapid response planning efforts (Objective IV,3).

a. Develop specific protocols for critical habitat areas of endangered and threatened species, marine sanctuaries, and estuarine reserves to minimize disruption of these areas. Prioritize efforts based on geographic proximity to areas already invaded.

3. Employ Geographic Information System (GIS) technology to refine the current and potential range of *C. taxifolia* in U.S. waters and prioritize surveillance efforts.

4. Develop and implement sound inspection protocols to effectively monitor imported commodities and detect the presence of *C. taxifolia,* including shipments that arrive in the U.S. through alternative mailing, shipping, and delivery services.

a. Develop and employ inspection protocols at all domestic ports of entry to monitor compliance with federal regulations regarding the importation of *C. taxifolia*.

OBJECTIVE IV -- ERADICATION

Eradicate populations of *Caulerpa taxifolia* already introduced into U.S. waters, and maximize "control readiness" to effectively implement eradication programs when new populations are discovered.

Problem

Like most invasive species, if *C. taxifolia* becomes established and widespread, control or complete eradication may not be possible. However, if detected early, there may be a brief window of opportunity to control and possibly eradicate small isolated populations. Successful eradication is therefore primarily dependent on early detection and "control readiness". It is imperative to develop effective eradication techniques that can be implemented quickly and identify federal and state regulatory and administrative leadership to ensure thorough and efficient handling of required permits and other documentation. Most states are not well prepared for such an invasion and likely would miss the narrow window for successful eradication.

The discovery of *C. taxiolia* in California and the eradication efforts that were instituted demonstrate the significance and effectiveness of these factors, providing a model for rapid response planning. *C. taxifolia* was discovered by an alert biologist during routine monitoring surveys. Within days, the identity of a collected specimen was confirmed, additional specimen were forwarded to European experts for genetic confirmation, state and federal agencies were alerted, and the first of many multi-agency eradication planning meetings was held. Within less than one month, field operations were underway to eradicate *C. taxifolia*. Many factors contributed to the state's rapid response including an awareness among federal and state agencies of the potential threats posed by this invasive species as well as the state's knowledge of regulatory and administrative requirements and environmental issues based on experience with other pest management and eradication programs. The California experience serves as a model of the coordinated, multi-agency approach needed to guide and implement eradication efforts.

Recommended Actions

1. Apply Integrated Pest Management principles to develop, research, and implement *C. taxifolia* control and eradication strategies including physical, chemical, and biological alternatives that are cost effective, and minimize the risk of harm to the environment and to human health and welfare. The availability of various control strategies will allow for flexibility and enhance the likelihood of execution when new populations are detected.

a. Identify potential control alternatives for consideration in critical habitat areas for endangered and threatened species, marine sanctuaries, and estuarine reserves.

b. Share research findings and control results with international partners to reduce populations at important ports of origin and with the Technical Advisory Committee developing rapid response eradication recommendations (see Recommended Action 3.a. below).

2. Conduct a scientific review of the rapid response eradication strategies implemented in California. Through a panel of experts, in both *C. taxifolia* and control, develop rapid response recommendations to prepare for future discoveries of *C. taxifolia* in U.S. waters.

3. Encourage and lead rapid response planning to foster "control readiness". Prioritize effort in areas where *C. taxifolia* has already been introduced and where dispersal is most likely to occur based on already established invasions or high risk pathways of introduction and spread.

a. Establish a Technical Advisory Committee for Rapid Response Assessment. The Committee, made up of *C. taxifolia* and control experts, will be called upon when a new population of *C. taxifolia* is detected to review all information gathered by local agencies, including pertinent habitat information, and provide recommendations for eradication and prevention to appropriate jurisdictional agencies.

b. Develop and use education and outreach strategies to inform state agencies with regulatory authority about rapid response planning and more specifically the role of the Technical Advisory Committee.

c. Identify lead agencies (federal and state) responsible for invasive species control in marine systems, and other agencies with jurisdictional interests in coastal states vulnerable to *C. taxifolia* invasion. Develop a rapid response call-sheet for each state providing current contact information for reporting and response. Ensure this information is shared with appropriate state and federal agencies, as well as other organizations and the public to facilitate efficient reporting.

d. Establish a federal funding mechanism for rapid response eradication efforts. Ensure adequate resources (minimum $100,000) are available and can be allocated immediately (10 business days) if *C. taxifolia* is found and confirmed in a new location.

4. Conduct an economic assessment analyzing the cost of control and prevention strategies relative to the potential cost of introduction and establishment of *C. taxifolia*. Incorporate various scenarios or levels of control, eradication, and prevention implementation (e.g. no action versus complete eradication and prevention). Use the California experience as a demonstration.

OBJECTIVE V -- RESEARCH

Support research initiatives to prevent the introduction, establishment, and dispersal of *Caulerpa taxifolia* in U.S. waters.

Problem

The prevention objectives and actions recommended in this document are based on the current state of knowledge. Current information gaps limit the scope and potential effectiveness of this Prevention Program. Additional knowledge gained through research is critical to the successful prevention of *C. taxifolia* infestations in U.S. waters. The refinement, update, and execution of this Prevention Program should reflect the information gained through future research efforts.

Research initiatives that advance the implementation of this Prevention Program are a priority. The identification of critical information needs and gaps will assist funding agencies in justifying and prioritizing proposed initiatives. Currently, field research in the U.S. is limited as the risk of dispersal is too high and the urgency for eradication is paramount. These limitations emphasize the need to work with researchers and resource managers from areas around the world that have been invaded by *C. taxifolia* (i.e. Europe and Australia) and in areas that host native *C. taxifolia* populations.

Recommended Actions

1. Identify researchers, managers, and administrators from universities, government agencies, and public and private organizations with expertise in *C. taxifolia* biology, ecology, and control to consult with the *Caulerpa taxifolia* Prevention Committee.

2. Conduct a thorough literature search to identify important information gaps, controversies, and conclusions.

3. Support research and monitoring initiatives that assess the viability of *C. taxifolia* plant fragments transported by various pathways including ballast water tanks, imported commodities, and attached to fishing and boating equipment and anchors. Determine threshold parameters for survival, if appropriate (e.g. time, temperature, relative humidity). Apply findings to risk analyses.

4. Support research and monitoring initiatives that focus on the direct and indirect ecological and economic impacts of the invasion and potential spread of *C. taxifolia* in U.S. coastal waters, environmental tolerances of *C. taxifolia* to advance range predictions, and the efficacy of short- and long-term eradication and control. Apply findings to risk analyses.

5. Establish an International Consortium of *Caulerpa* Researchers to advance research initiatives, especially field programs that can not be completed in U.S. coastal waters, develop research protocols to standardize research methods globally, and disseminate research results. Explore potential funding mechanisms to support such a Consortium. Host an annual meeting of the Consortium to ensure information sharing.

6. Identify potential funding sources for eradication efforts, research and monitoring programs, and workshops.

OBJECTIVE VI -- REGULATION

Support efforts to enforce existing federal and state regulations and encourage compliance to prevent the introduction and dispersal of *Caulerpa taxifolia*.

Problem

In March 1999, the U.S. Department of Agriculture promulgated regulations under the Federal Noxious Weed Act of 1974 (subsequently replaced by the Plant Protection Act) restricting and requiring permits for the importation of the Mediterranean clone of *C. taxifolia* (Appendix G). However, resource constraints and the inability to distinguish the invasive Mediterranean strain from other un-listed *Caulerpa* species present challenges to the enforcement of these regulations. APHIS may, however regulate import or interstate movement of any taxon not readily distinguishable from *C. taxifolia*.

Preventing the introduction of *C. taxifolia* in commercial imports will require the cooperation of federal and state inspection and enforcement agencies. Federal agencies likely to contribute to *C. taxifolia* prevention through enforcement responsibilities include the Food and Drug Administration (FDA), NOAA, the Service, U.S. Customs Service, and APHIS. The State of California is the only state that has responded to the threat of *C. taxifolia* with regulatory action. On September 25, 2001, regulations prohibiting the importation, possession, transport, transfer, sale or giving away of, and release of nine species of *Caulerpa* were enacted. All nine species are either difficult to distinguish from *C. taxifolia* or are considered to be potential invaders. Although these regulations provide the state with the necessary authority to confiscate and destroy these species, the lack of funds for personnel has restricted enforcement capabilities. The City of San Diego, CA prohibits the sale and possession of all *Caulerpa* species (Chapter 6 Article 3 Division 15 of San Diego Municipal Code) (Appendix F). This represents the most comprehensive ban of *Caulerpa* species, demonstrating the commitment of the city to protect its coastal resources from invasive *Caulerpa* species.

Education initiatives, cooperation, and consistent enforcement are critical to successfully preventing *C. taxifolia* introductions through commercial imports.

Recommended Actions

1. Develop inspection and enforcement protocols, emphasizing cooperation and consistency among inspection agencies to prevent the introduction of *C. taxifolia* though commercial imports. Use education and outreach strategies to advance awareness among enforcement agents about legislative authorities and to achieve consistent enforcement.

a. Develop protocols that are pathway specific to focus efforts and resources.

b. Define reporting requirements and expectations of enforcement agents to ensure efficient reporting and confiscation in the event that *C. taxifolia* is detected.

c. Organize multi-agency workshops and training to foster cooperation among federal agencies and state agencies.

d. Address holding, treatment, and disposal of products containing *C. taxifolia*.

2. Support package labeling requirements that report all contents of packaged goods, including packing materials, to maximize inspection efficiency.

3. Develop enforcement agency programs to monitor or evaluate industry compliance with regulations, including permitting and labeling requirements.

4. Develop partnerships and cooperative agreements with foreign governments to estimate and regulate the use of *C. taxifolia* in commercial shipments exported to the U.S.

a. Collaborate with foreign governments and industry representatives to establish a "*Caulerpa* Free" Certification Program for all imports containing aquatic materials. The program will foster compliance with regulatory authorities.

OBJECTIVE VII -- LEGISLATION

Support the use and development of consistent legislative authorities to prevent the introduction, establishment, and dispersal of *Caulerpa taxifolia* in U.S. waters.

Problem

Existing legislative authorities available to resource and enforcement agencies to prevent the introduction and spread of *C. taxifolia* are limited in scope. The Plant Protection Act is the only existing federal authority that regulates the movement of *C. taxifolia* into the U.S. (Appendix D). Although there are provisions regarding interstate transport within the Act, there are no restrictions on intrastate movement. The State of California is the only state that has enacted legislation to regulate the movement of *C. taxifolia* within state boundaries. Preventing the introduction and spread of *C. taxifolia* in the U.S. will require consistent and enforceable federal and state legislative authorities imposing strict and severe penalties for noncompliance.

Recommended Actions

1. Conduct a thorough review and analysis of existing and proposed legislative authorities to assess applicability to prevention activities and identify gaps.

2. Develop new legislative and regulatory initiatives to expand the scope of existing legislation and eliminate current gaps.

a. Implement a broader ban (genus-level, if necessary and supported by appropriate documentation including risk assessment and distribution analyses) on the importation of *Caulerpa*, except as permitted for research, to eliminate identification problems for enforcement agencies and aquarium enthusiasts (domestic and international) and to minimize the risk of introducing other species of non-native and potentially invasive *Caulerpa*.

3. Coordinate and guide state agencies to develop, enact, and enforce consistent state legislation and regulatory authorities.

a. Utilize existing multi-state organizations or commissions to advance regional policy, consensus, and commitment.

OBJECTIVE VIII -- EDUCATION

Increase awareness among all sectors of the public emphasizing the risks posed by *Caulerpa taxifolia* and the importance of and need for public participation and support in prevention activities.

Problem

Public support and cooperation is required to successfully implement any comprehensive prevention or control program. Public support has made significant contributions to detection, prevention, and control of other introduced, invasive species. However, without the continued exchange of timely, accurate and consistent information, the public can unknowingly contribute to the introduction, spread, and establishment of invasive species like *C. taxifolia*.

A comprehensive *C. taxifolia* educational initiative will foster an understanding of the risks associated with its introduction and spread. Entities likely to be impacted by or affecting the spread of *C. taxifolia* require knowledge and awareness to incorporate appropriate prevention actions into routine activities and ensure compliance with regulatory authorities.

Recommended Actions

1. Develop *C. taxifolia* educational and outreach initiatives targeting general and specific audiences. Utilize existing programs, agencies, or multi-state organizations to facilitate efficient information transfer and distribution of educational materials.

a. Identify specific audiences or entities potentially affecting or impacted by the introduction and spread of *C. taxifolia* or the implementation of prevention activities. Target groups include, but are not limited to:

i. Aquarium industry (personal and commercial hobbyist, wholesalers, retailers, and trade groups)
ii. Zoological parks and museum associations
iii. Coastal property owners and marinas
iv. Commercial and recreational fishery
v. Boaters and personal watercraft users
vi. Bait industry (personal and commercial harvest, retailers, wholesalers)
vii. Divers
viii. Government officials (including enforcement agents) and policy-makers
ix. General public

2. Encourage aquariums, zoos, museums and other public facilities using aquatic organism displays to eliminate live *C. taxifolia* from all displays and use alternative native species. Develop invasive species displays that focus on the overall risks posed by invasive species, including *C. taxifolia*, and feature basic prevention strategies, regulations, and the importance of public participation.

3. Coordinate and establish effective information sharing among scientists, government agencies, and the public to promote prevention activities. Establish an information clearinghouse to maintain and disseminate information. Establish a clearinghouse web site to ensure Internet accessibility.

LITERATURE CITED

Anderson, L.W.J. and S.M. Keppner. 2001a. *Caulerpa taxifolia*: Marine algal invader provokes quick response in U.S. waters. Aquatic Nuisance Species Digest 4(2):13, 21-23.

Anderson, L.W.J. and S.M. Keppner. 2001b. *Caulerpa taxifolia*: New marine algal invader in U.S. waters. Aquatic Invaders: the Digest of the National Aquatic Nuisance Species Clearinghouse 12(1):1-6.

Bellan-Santini, D., P.M. Arnaud, G. Bellan, and M Verlaque. 1996. The influence of the introduced tropical alga *Caulerpa taxifolia*, on the biodiversity of the Mediterranean marine biota. Journal of Marine Biology Assoc UK 76:235-237.

Boudouresque, C.-F., A. Meinesz, M. Verlaque, and M. Knoeffler-Peguy. 1992. The expansion of the tropical alga *Caulerpa taxifolia* (Chlorophyta) in the Mediterranean. Cryptogamie Algologie 13:144-145.

Ceccherelli, G., and F. Cinelli.1998. Habitat effect on spatio-temporal variability in size and density of the introduced alga *Caulerpa taxifolia*. Marine Ecology Progress Series 163:289-294.

Ferrer, E., A. Gomez Garreta, and M. Antonia Ribera. 1997. Effect of *Caulerpa taxifolia* on the productivity of two Mediterranean macrophytes. Marine Ecology Progress Series 149:279-287.

Francour, P., M. Harmelin-Vivien, J.G Harmelin, and J. Duclerc. 1995. Impact of *Caulerpa taxifolia* colonization on the littoral ichthyofauna of Northwestern Mediterranean Sea: preliminary results. Hydrobiologia. 300/301:345-353.

Giannotti, A., E. Ghelardhi, and S. Sinesi. 1994. Characterization of seawater bacterial communities within environments colonized by the tropical green seaweed *Caulerpa taxifolia*. *In* Boudouresque, C.-F., A. Meinesz, and V. Gravez (eds). First International Workshop on *Caulerpa taxifolia*. GIS Posidonie Publications, Marseille, France. pp. 197-201.

Guerriero, A., A. Meinesz, M. D'Ambrosio, F. Pietra. 1992. Isolation of toxic and potentially toxic sesqui-and monoterpenes from the tropical green seaweed *Caulerpa taxifolia* which has invaded the region of Cap Martin and Monaco. Helv Chim Acta 75:689-695.

Harmelin-Vivien, M., J.G. Harmelin, and P. Francour. 1996. A 3-year study of the littoral fish fauna of sites colonized by *Caulerpa taxifolia* in the N.W. Mediterranean (Menton, France). In: Ribera, M.A., E. Ballesteros, C.F. Boudouresque, A. Gomez, and V. Gravez (eds). Second International Workshop on *Caulerpa taxi*folia. Publicacions Universitat Barcelona. pp 391-397.

Jousson, O., J. Pawlowski, L. Zaninetti, A. Meinesz, and C.F. Boudouresque. 1998. Molecular evidence for the aquarium origin of the green alga *Caulerpa taxifolia* introduced to the Mediterranean Sea. Marine Ecology Progress Series 172:275-280.

Keppner, S.M. and R.T. Caplen. 1999. *A Prevention Program for the Mediterranean Strain of Caulerpa taxifolia*. U.S. Fish and Wildlife Service, Lower Great Lakes Fishery Resources Office, Amherst, NY.

Keppner, S.M., M.T. Weimer, and W.J. Troxel. 1998. *Caulerpa taxifolia: A Potential Threat to U.S. Coastal Waters*. U.S. Fish and Wildlife Service, Lower Great Lakes Fishery Resources Office, Amherst, NY.

Komatsu, T., A. Meinesz, and D. Buckles. 1997. Temperature and light responses of alga *Caulerpa taxifolia* introduced into the Mediterranean Sea. Marine Ecology Progress Series 146:145-153.

Meinesz, A.(translated by D. Simberloff) 1999. Killer Algae. The University of Chicago Press. Chicago, IL. 360p.

Meinesz, A., J. de Vaugelas, B. Hesse, and X. Mari. 1993. Spread of the introduced tropical green alga *Caulerpa taxifolia* in northern Mediterranean waters. J Appl Phycol 5:141:147.

Merino, V., E. Clavero, and M. Hernández-Mariné. 1994. The effect of *Caulerpa taxifolia* (Vahl) C. Agardh on the growth of marine microalgae. *In* Boudouresque, C.-F., A. Meinesz, and V. Gravez (eds). First International Workshop on *Caulerpa taxifolia*. GIS Posidonie Publications, Marseille, France. pp.213-217.

Olsen, J.L., I. Meusnier, W.T. Stam, and M. Valero. 2002. Tracing invasions with genetic markers. International *Caulerpa taxifolia* Conference, San Diego, CA. January 31, 2002.

Raloff, J. 1998. Rogue algae. Science News Online. from Science News, Vol. 154, No. 1, July 4, 1998, p.8.

Appendix B. Contributors to the draft National Management Plan for the Genus *Caulerpa*, 2004.

Caulerpa Working Group Members in 2004

Name	Organization
Alcocer, P. Rick	U.S. Department of Homeland Security
Anderson, Lars	U.S. Department of Agriculture, Animal and Plan Health Inspection Service
Bell, Carl	National Oceanic and Atmospheric Administration, SeaGrant
Bergendorf, David	U.S. Fish and Wildlife Service
Brockbank, Marcia	San Francisco Estuary Porject/Association of Bay Area Governments
Cravalho, Domingo	HI Department of Agriculture
Ellis, Susan	CA Department of Fish and Game
Gulkok, David	HI Dept. of Land and Natural Resources
Heimowitz, Paul	U.S. Fish and Wildlife Service
Hoffman, Robert	National Marine Fisheries Service
Kenworthy, Jud	National Oceanic and Atmospheric Administration
Kumpf, Herb	National Oceanic and Atmospheric Administration
Lapointe, Brian	Harbor Branch Oceanographic Institution
Leavitt, Robert	CA Department of Food and Agriculture
Lehtonen, Polly	U.S. Department of Agriculture, Animal and Plan Health Inspection Service
Lonhart, Steve	Monterey Bay Aquarium
McDowell, Karen	San Francisco Estuary Porject/Association of Bay Area Governments
Meyers, Marshall	Pet Industry Joint Advisory Council
Murray, Steven	CA State, Fullerton
Posthumus, Bruce	San Diego Regional Water Quality Control Board
Tasker, Al	U.S. Department of Agriculture, Animal and Plan Health Inspection Service
Torbett, Tim	U.S. Departmento of Agriculture
Walters, Linda	University of Central Florida
Williams, Erin	U.S. Fish and Wildlife Service
Williams, Susan	UC, Davis

Other Contributors in 2004	
Name	**Organization**
Hardin, Scott	Florida Fish and Wildlife Conservation Commissioin
Madl, Pierre	Independent contributor
Millar, Alan	Royal Botanic Gardens Sydney
Moore, Jim	California Department of Fish and Game
Peterson, Jenny	U.S. Fish and Wildlife Service
Peyton, Kim	University of Hawaii
Silva, Paul	University of California at Berkeley
Squires, Kim	U.S. Fish and Wildlife Service
Woodfield, Rachel	Merkel and Associates, Inc.
Yip, Marcela	Independent contributor

Appendix C. Literature Review of the Aquarium Strain of *Caulerpa taxifolia*

Appendix C. Literature Review of the Aquarium Strain of *Caulerpa taxifolia*

Prepared by: Pierre MADL and Maricela YIP
University of Salzburg - Faculty of Molecular Biology,
Hellbrunnerstrasse. #34, A-5020
Salzburg Austria

Revised July, 2005

Available on the internet at: http://www.sbg.ac.at/ipk/avstudio/pierofun/ct/caulerpa.htm

Table of Contents:

1.a Introduction

In the early 1980s, the curator of the tropical saltwater aquarium at the *Wilhelmina* Zoo in Stuttgart (FRG), noticed the exceptional properties of a bright green, beautiful green alga, *Caulerpa taxifolia*, used as tank decoration in the presentation of multicolored tropical fish. It was captively bred by the aquarium staff and exposed, for years, to chemicals and ultraviolet light. This exposure to abiotic stressors altered and switched on genes that have not been previously present, expressed or active in wild type strains found across the Pacific. The genetically altered seaweed, in contrast to other algae does not wither and grows with astounding vigor resisting cool water temperatures. Specialists quickly learned about these qualities, and public aquaria around the globe acquired cuttings. Eventually, in 1982, a sample of it found its way to the Oceanographic Museum in *Monaco*, then headed by *Jacques Cousteau*. Two years later marine biologist *Alexandre Meinesz* discovered the alga in nature, under the windows of this celebrated building.

Since then the alga has colonized huge areas and is still progressing unchecked into new habitats in unstoppable manner. The algae grows everywhere, from the surface to the lower limits of underwater vegetation to capes swept by storms and currents, on soft bottoms of sheltered bays, on polluted mud of harbors as well as on stretches of bottom with a diverse flora and fauna. With its highly toxic defense cocktail, it is barely devoured by herbivores, thus facilitating its limitless spread. It is thus growing unrestrained, covering and then eliminating many autochthonous plant and animal species. A new ecological equilibrium is reached once the alga forms a dense, uniform carpet that persists from year to year. Hence, the Global Invasive Species Specialist Group categorized *C.taxifolia* among the 100 most "Worst Invasive Alien Species" threatening biodiversity ([1.0]ISSG, 2004). This alga has become the sort of "evil plant" that is well reflected in *Charles Baudelaire*'s poem:

Both of you are discreet, dim, shadow-ridden:
Man, none has plumbed your soul's abyss; and, sea,
No one has pierced your wealth's dark mystery,
So jealous, you, to keep your treasures hidden!

Not surprisingly it acquired negative fame as the "Aquarium-Mediterranean strain" ([1.1]Jousson et al., 2000) or even publicized as the "Killer Algae" ([1.2]Meinesz, 1999). This is underlined by recent discoveries of *Caulerpa taxifolia* at the coast of California (USA) and New South Wales (AUS) raising public concern about the potential danger of a new invasion similar to the one endured by the Mediterranean Sea over the past decades.

Fig.1.a: Oceanographic Museum of Monaco.

1.b Historical Flashbacks

The chronology of the Mediterranean invasion of *C.taxifolia* reads like a tale of missed chances and spoiled opportunities:

1984: a few meters deep, A.Meinesz, professor at the University of *Nice-Sophia Antipolis*, localizes a patch of *C.taxifolia* about a $1m^2$ in size situated just at the base of the Oceanographic Museum in *Monaco*.

1989: repeated surveys reveal that the original site of infestation extends to cover an area about 1ha in size.

1990: in summer of that year, the alga has spread along the southeastern coast of France. First outcrops are also spotted some 100km west of *Monaco* at *Cap Martin* and *Toulon*.

1991: by now the radius of infestation stretches as far as 400km west of Monaco at *St.Cyprien* close to the Spanish border.

1992: early that year and for the first time, the alga is spotted at sites opposing the westward drifting Ligurian current: e.g. *Imperia* (Italy); on the western front it continues its spread westward to hit the coast of the *Balearic* Island of *Majorca* (Spain).

1993: the southeastward extension of the algae continues; sites of infestations are registered at the island of *Elba* (Italy) and as far south at *Messina* in the strait of Sicily (Italy).

1995: eventually the alga reaches the Adriatic Sea; first sightings are reported at *Malinksa* and *Stari Grad*

(Croatia).

1997: by the end of that year, the tumor-like growth of individual patches of *C.taxifolia* carpets along the south-eastern coast of France have drastically increased in size and eventually merge to form huge monoclonal meadows.

2000: in ever more rapid events, *C.taxifolia* reaches the southern shores of the Mediterranean and is sighted at *Sousse* (Tunesia). In the very same year a large patch of this invasive species is identified in the lagoon off *San Diego* (California, USA). At the same time patches of this invasive strain are sighted in *Port Hacking*, *Lake Conjola* and *Careel Bay* (in New-South Wales, AUS).

2001: eventually other spots of infestation are identified at sites along the south-eastern coast of Australia: e.g. *Narrawallee Inlet*, *Botany Bay*, *Burrill Lake*, *Lake Conjola* and *Berringer Lake*, *Careel Bay*, *Pittwater*, *Lake Macquarie* all in New South Wales and *West Lakes & Port River* in South Australia.

2002: in April of that year, significant infestations of *C.taxifolia* are documented in the northern part of *Sydney harbour* and later that year in *St.Georges basin* (120km south of *Sydney*) - again in NSW, Australia ([1.2b]Creese et al., 2004).

Fig.1.b: Spread of *C.taxifolia* along the French coast.

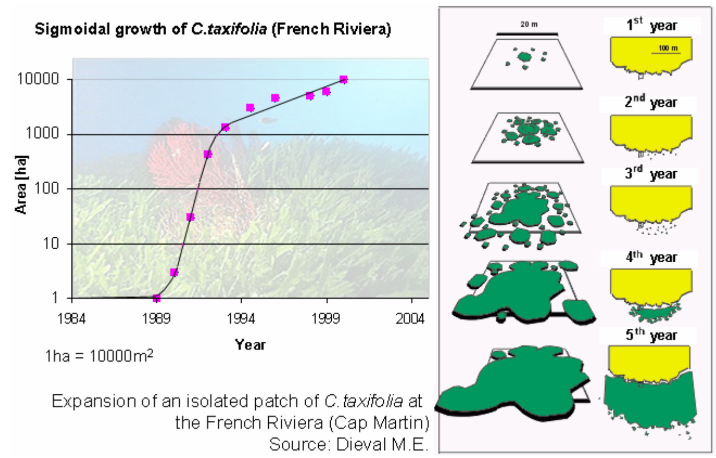


Fig.1.c: C.taxifolia in the W-Mediterranean.

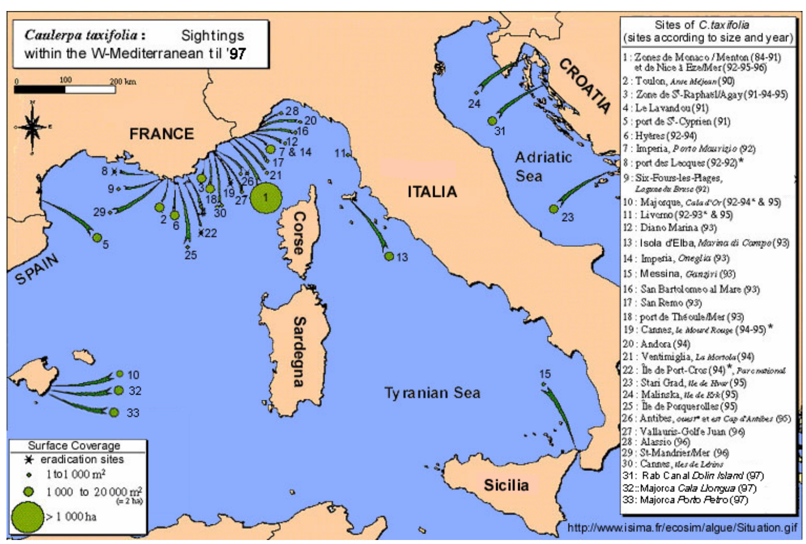


Fig.1.d: *C.taxifolia* on a global scale.

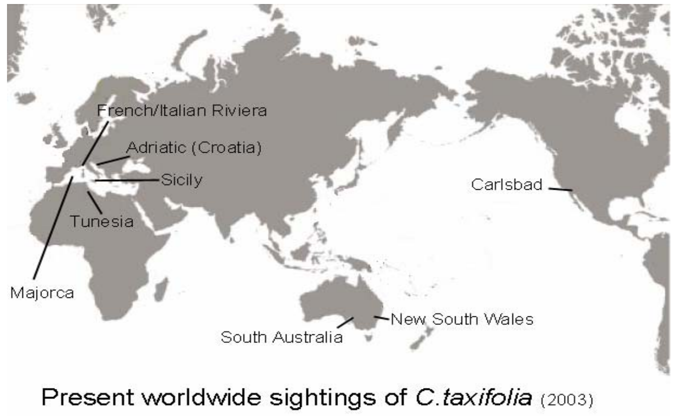


Fig.1.e: Global distribution of native *C.taxifolia* ([1.2a]Nimpis, 2002).

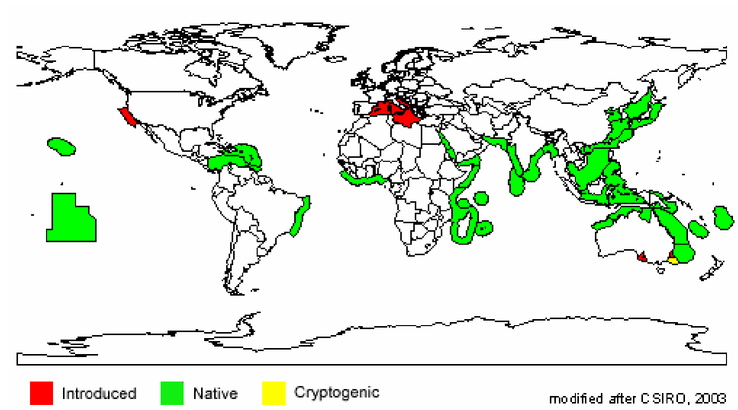
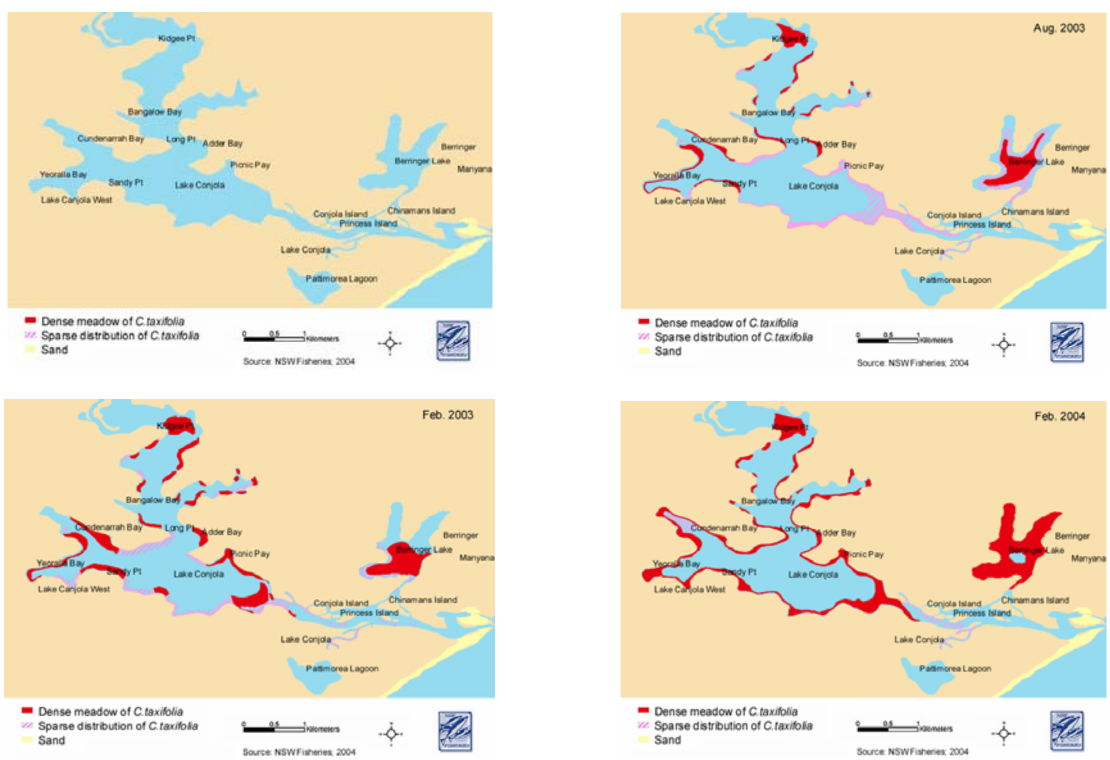


Fig.1.f: *C.taxifolia* in *L.Canjola*, NSW - AUS (below).



Prolific growth of *Caulerpa* along the *Cote d'Azur* (France), where the introduction was first reported, has been associated with urban wastewater pollution ([1.3]Chisholm et al., 1997). It easily proliferates vegetatively via fragmentation aided by subsequent dispersal via anchors and fishing nets ([1.4]Meinesz, 1992), or dumping ballast water across the oceans; in particular at harbors, marinas and other places where boats anchor ([1.6]Boudouresque et al., 1995). Mid range spread of this species is easily achieved by currents, which transport fragments of it into new areas yet to be colonized ([1.3]Chisholm et a., 1997). Apart from shipping vectors, long range dispersal of this alga was facilitated by the aquarium trade ([1.7]Schaffelke et al., 2002). The fact that *C.taxifolia* possess a chemical defense mechanism (the alga produces repellent toxins) renders it unpalatable to generalist herbivores in the N-W Mediterranean ([1.8]Paul, 2002), which facilitated this biological invasion. Thus, *C.taxifolia* is upsetting the biocoenosis by invading and out-competing the indigenous flora while protecting itself against predation, thus threatening the biological stability of the marine environment ([1.9]Pesando et al., 1996). Apart from a few serious attempts to eradicate this species (mainly in AUS and USA), monitoring, mapping and public awareness programs are the only efforts made so far. It seems that control of the invasion was and still is never a priority for most of the affected EU-countries. A shameful attitude that aids in the dispersal of this invasive strain.

Fig.1.g: *C.taxifolia* meadow of the Mediterranean.

2. Phylogeny, Morphological & Genetic Features

2.a Phylogeny of *C.taxifolia*: The tropical strain of *Caulerpa taxifolia* is widespread among the tropics; with native populations found in the Atlantic Ocean (West Indies and African coast), the Indian Ocean (Pakistan, Sri Lanka, and north western Australia), and the Pacific Ocean (Philippines, Indonesia, Japan, New Caledonia, and north eastern Australia) where it grows in small patches and does not present problems. This marine, green alga, was first described by *M.Vahl* in 1802 as *Fucus taxifolius* and was regrouped in 1817 by *C.Agardh* into the following taxonomic position:

KINGDOM **Protista** (Gk. protos, first) by Raven et al., 1992 or
Plantae (L. plant); by Postlethwait & Hopson, 1995 & [2.0]ITIS, 2004;
DIVISION / PHYLUM **Chlorophyta** (Gk. chloros, green + phyto, plant);
CLASS **Ulvophycea** (L. ulva, sedge + phyto, plant);
Chlorophyceae according to [2.0]ITIS, 2004;
ORDER **Caulerpales** (L. caulis, stalk) referring to the stalk of the algae;
Bryopsidales (unicellular & multinucleate thalli) according to [2.0]ITIS, 2004;
FAMILY **Caulerpaceae** (L. caulis, stalk);
GENUS ***Caulerpa*** (L. caulis, stalk);
SPECIES ***taxifolia*** (L. taxi, yew-like + folia, leaves) referring to the dark-green and flattened foliage;

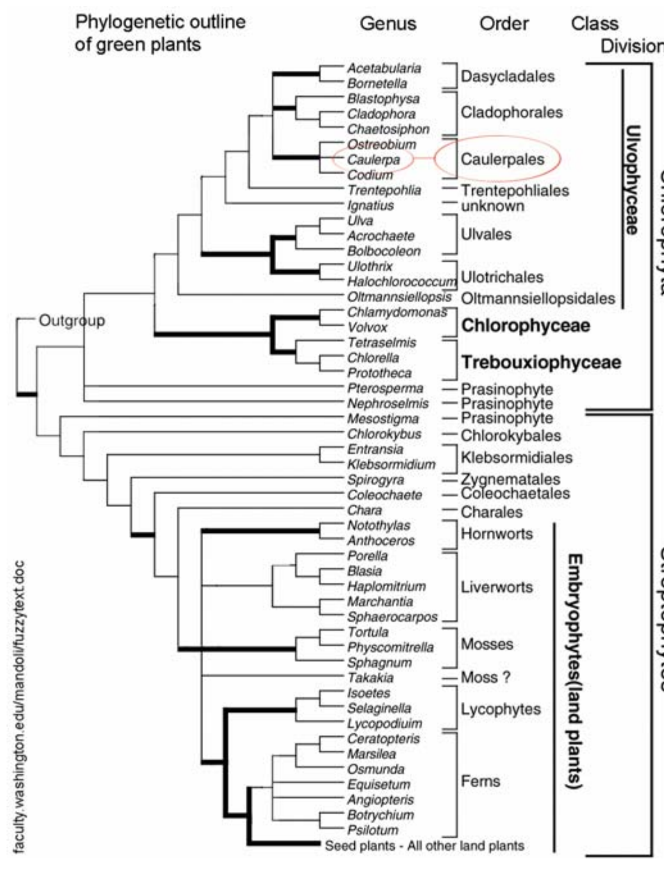


Fig.2.a: Taxonomic position of *C.taxifolia*.

The **Chlorophyta** (green algae) groups unicellular or multicellular photosynthetic organisms characterized by the presence of chlorophyll a and b, as well as various carotenoids. The carbohydrate food reserve is starch, which is stored in plastids. There are about 7000 known species ([2.1]Raven et al., 1992).

The **Ulvophyceae** is one of the 3 classes currently recognized in the chlorophyte lineage, the others being Chlorophyceae and Trebouxiophyceae (formerly Pleurastrophyceae) and is made up of approximately 1100 species ([2.2]Dumay et al., 2002) clustered among 100 genera. Most of which are found in temperate and tropical marine environments, and the majority of green "seaweeds", including well-known species of besides *Ulva* and *Acetabularia* besides *Caulerpa*, are placed in this class as well. Conversely, the Chlorophyceae and Trebouxiophyceae, and the Streptophytes, consist almost entirely of non-marine organisms. In classifications based on morphology and ultrastructure, the Ulvophyceae have been separated from other chlorophytes mostly on the basis of characters associated with mitosis, cytokinesis, and the flagellar apparatus of zoospores and gametes. In molecular analyses, however, the relationships among these three classes are less clear ([2.3]Mandoli et al., 2002). Moreover, the "siphonous" orders of Ulvophyceae (Cladophorales, Dasycladales, Caulerpales) are difficult to resolve vis-à-vis each other and with other Ulvophyceae (orders Ulotrichales and Ulvales); phylogenetic trees based on single gene sequences reveal long branch lengths between "siphonous" sequences and those of other chlorophytes ([2.3]Mandoli et al., 2002).

Members of the order **Caulerpales** contain marine (sub-) tropical macrophytes with multinucleate siphonous forms, without cross-walls or segregative division, filled with numerous plastids (e.g. amyloplasts which is an unpigmented plastids for starch storage). Thus forming one huge cell, which is vulnerable to substantial plasma loss. Caulerpales though, are equipped with efficient wound healing properties (upon damage, healing occurs in seconds, involving actin-mediated contraction and a "plug" of cell wall material). The skeletal constituent of the cell wall contains xylan rather than cellulose and is reinforced with a multiaxial construction, and sometimes even stabilized with calcium carbonate deposits as in *Halimeda* sp.([2.4]Strassburger, 1998).

The thallus among members of the genus ***Caulerpa*** - in which approximately 75 species are currently recognized - is non-septated but siphonous in structure and reinforced with anastomosing strands of wall material (trabeculae) which are ingrowths of the cell wall. A thin layer of cytoplasm, containing countless numbers of each type of organelles, is appressed to the wall ([2.5]Silva, 2002). Division of labor is achieved between photosynthetic chloroplasts and starch-storing leucoplasts (heteroplastidy).

Furthermore, *Caulerpa* possess siphonaxanthin and siphonein as photosynthetically active pigments ([2.5]Silva, 2002). This alga also exhibits some particular biological and physiological features: resistance to low temperature, gigantism of the thallus, high growth rate, which differentiate it morphologically from the tropical strain ([2.6]Meinesz et al., 1995).

2.b Morphology of *C.taxifolia*: Although individual plants are composed of only one cell, *Caulerpa* has a complex morphology, composed of pseudo-organs that often resemble the roots, shoots and leaves of higher plants. It is one of the most distinctive genera of seaweeds, making it identifiable solely on the basis of its habit ([2.5]Silva, 2002). It consists of a creeping rhizome that produces tufts of colorless rhizoids downward and photosynthetic branches (assimilators) upward. When dealing with dimensions, the aquarium strain is different in size, length, growth rate and temperature tolerance from samples collected in tropical areas ([2.7]Boudouresque et al, 1995). *C.taxifolia* in the coastal areas of tropical North to Central QLD and from *Port Hacking* has a delicate morphology with narrow stolons, fronds, and pinnules ([2.8]Benzie et al., 2000). In contrast, the aquarium strain of *C.taxifolia* in the Mediterranean is generally quite large with broader stolons, fronds, and pinnules, although variable depending on depth (larger in low light conditions) and season ([2.6]Meinesz et al., 1995). However, populations of *C.taxifolia* in southern Queensland, at *Stradbroke Island*, which experience environmental conditions more akin with the Mediterranean, express a range of morphologies that include plants with heavy stolons and long, broad fronds bearing large pinnules ([2.8]Benzie et al., 2000).

Fronds: One of the more striking external features regards the dimensions of the vertical fronds or pinnae (L. feather, the large but divided leaf - see figure 2.b). Fronds may be quite short or even absent in shallower water (leaving only the stolons), but are longer in deeper water with prevailing low light conditions. In the tropical version, primary fronds are 2-15cm in length, while those of the Mediterranean strain range reach average frond heights of 25cm - ranging from 5cm in shallower water, to 40cm at depths of 15m, and even to 60-80cm at greater depths ([2.6]Meinesz, 1995). The increase in frond length, under conditions of competition with *Posidonia oceanica* likewise indicates the alga's property of adaptation ([2.2]Dumay et al., 2002). Starting from the base, each frond is made up of the midrib or rachis (Gk. backbone), which represents the prominent central axis. Frond density varies according to the season and ranges from 5100/m^2 in September to 14000/m^2 in April ([2.6]Meinesz et al., 1995).

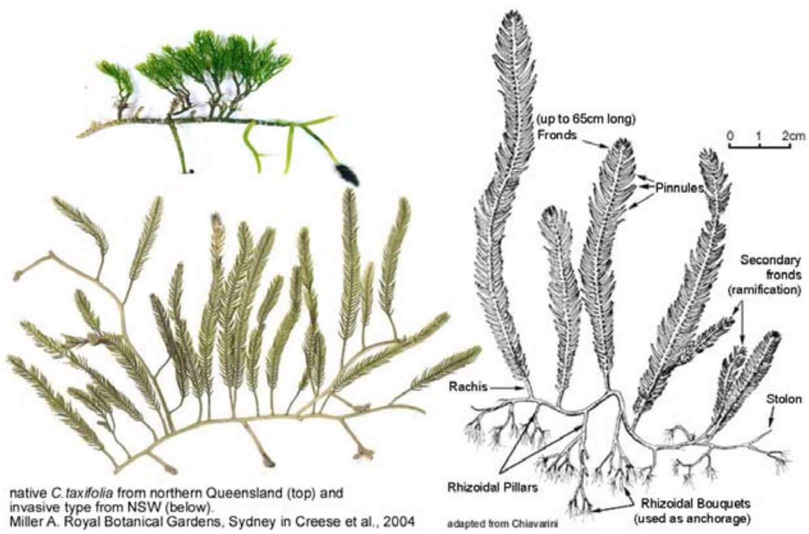
Rachis (Gk. backbone) is the main axis of the leaf (frond), from which the pinnules arise.

Pinnules (L. small feathers) grow out of this midrib to give each frond the characteristic feather-like appearance. Ramificating or branching fronds originate from primary pinnules. Pinnule length typically measures 1cm while pinnule density per side varies from 4 to 7/cm of rachis length. The shape of the pinnules is usually up-curved, tapered at the ends, with some even bifurcated at the top ends (split in two). According to Meinesz ([2.10]1995), pinnule spacing and length is light dependant.

Stolon (L. shoot): Regularly spaced fronds are attached to the horizontally running stolon and is also the origin of the adventitious rootless or rhizoids. After the winter, regrowth originates from old stolons that have survived - maximum stolon length is 2.8m, while the overall cumulative stolon length tends to stabilize around an equilibrium value of $230m/m^2$ ([2.11]Meinesz & Hesse, 1991).

Rhizoids (Gk. roots) are rootlike extensions that absorb water, food, and nutrients. Unlike in vascular plants, Caulerpales do not have a root system composed of root cap and root hairs. Instead, regularly spaced rhizoid pillars descend vertically from the stolons. A few centimeters long, these pillars branch up into extremely thin filamentous rhizoids, that based on the substrate, can form a felt-like net penetrating through the substrate and thus stabilizing it along with the algae ([2.10]Meinesz, 1995; [2.12] Chisholm et al., 1996).

Fig.2.b: Morphology of *C.taxifolia* [2.6a]Creese et al., 2004.



The debate whether *C.taxifolia* is an ecomorphic variant (an ecad) of another species, *C.mexicana* ([2.13]Jaasund, 1977; [2.14]Coppejans and Beekman, 1990; [2.15]Coppejans and Prud'homme van Reine, 1992), or a well-defined species in its own right ([2.16]Calvert et al., 1976; [2.17]Silva et al., 1987) is still unsolved. According to most accepted taxonomic criteria, ecomorphic variants of *C.taxifolia* can, under certain conditions, possess the morphological characteristics attributed to *C.mexicana*, and vice-versa ([2.18]Chisholm et al., 1995; [2.19]Olson et al., 1998). In order to differentiate *C.taxifolia* from the others, it must be mentioned that *C.taxifolia* is not the only invasive species of this genus that has conquered the Mediterranean Sea. While *C.taxifolia* has been accidentally released from the public aquarium at *Monaco*, *C.racemosa*, *C.scalpelliformis* and *C.mexicana* are considered to be Lessepsian immigrants from the Red Sea ([2.7]Boudouresque et al., 1996; [2.20]Piazzi et al., 1997).

Fig.2.c: In Situ morphology of *C.taxifolia*.

Therefore a brief excursion helps to highlight the morphological differences between *Caulerpa taxifolia*, *C.mexicana*, *C.scalpelliformis*, *C.racemosa* and the autochthonous species *C.prolifera*.

Caulerpa mexicana:
Partings of rhizoids are very closed on the stolon ((<1cm). The pinnules are wide and short; the ratio length on width (L/w) ranges from 2.5 to 5, and is a function based on environmental conditions. Fronds reach an average length of 6cm. Minimal viable temperature is 16°C.

Fig.2.d: *C.mexicana*.

Caulerpa scalpelliformis: Straight fronds that are about 10-20cm long. It closely resembles *C.taxifolia* but can be distinguished by the form of its branches, which are curved towards the interior in the latter and straight in the former. The rachis of the fronds is quite dominant and thicker than the pinnules. Another feature that distinguishes it from *C.taxifolia* are its pointed pinnules.

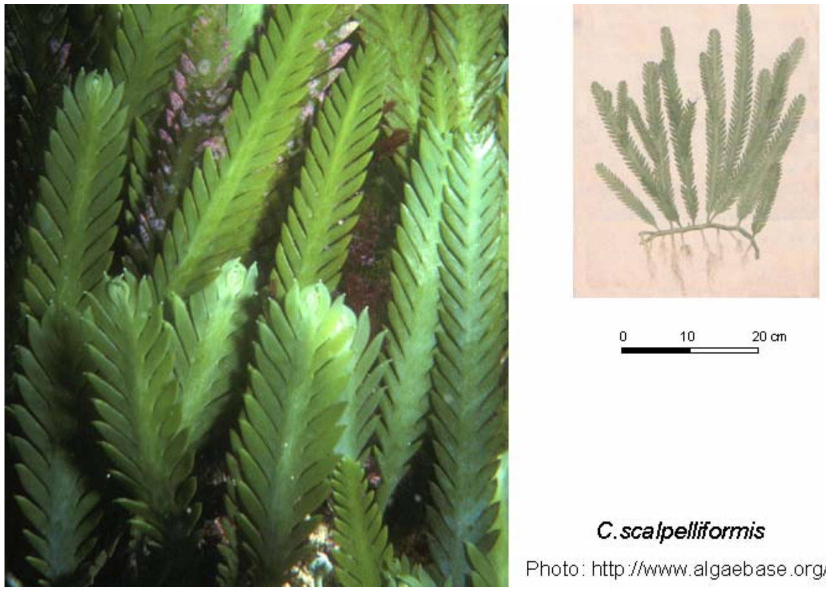


Fig.2.e: *C.scalpelliformis*.

Caulerpa racemosa: An algae with long stolons; its racemous-rhizoids point downward. Photosynthesis occurs in the clustered aggregations that, opposing the rhizoids, merely reach a few cm in height. Growth pattern of phtotosynthetic thalli can vary from bifurcated with tiny branches to entirely rounded bubbles.

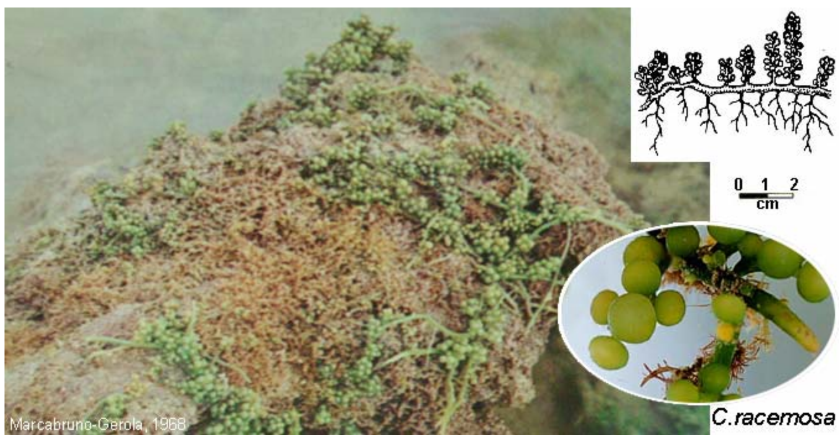


Fig.2.f:[2.37] *C.racemosa*.

This algae is likewise extremely invasive in Mediterranean subtidal habitats and as with *C.taxifolia* causes major modifications to benthic communities ([2.21]Verlaque & Fritayre, 1994; [2.22]Villele & Verlaque, 1995; [2.23]Bellan-Santini et al., 1996; [2.24]Ceccherelli & Cinelli, 1997; [2.25]Piazzi et al., 2001). It is currently in the process of spreading into the western Mediterranean ([2.26, 2.27]Piazzi et al., 1994, 1997; [2.28]Gambi & Terlizzi, 1998; [2.29]Modena et al., 2000; [2.30]Verlaque et al., 2000) and it seems to be capable of even out-competing *C.taxifolia* ([2.31]Ceccherelli et al., 2002).

Caulerpa prolifera: This native alga has few but robust stolons with the branching blades protruding perpendicularly into the water in intervals of 1-2cm. The superior part of each blades house the assimilatory pigments. The blades can reach a length of up to 15cm and can in some cases be even 13cm wide.

Fig.2.g: *C.prolifera*.

Usually they have an oval or linearly elongated appearance with a smooth border following a helical pattern towards the apex ([2.37] Marcabruno-Gerola, 1968).

2.c Genetic Features of *C.taxifolia*: The DNA fingerprints of *C.taxifolia* presented here support existing evidence for the descent of the Mediterranean *C.taxifolia* from an aquarium strain. The introduction of *C.taxifolia* via the Oceanographic aquarium in Monaco is strongly supported on the basis of having identical internal transcribed spacer (ITS) rDNA sequences ([2.32]Jousson et al., 1998). The phylogenetic analysis of these sequences show that the Mediterranean alga is genetically identical to the strain cultivated in aquaria (see figure 2.h - left image). Interestingly, the aquarium strain differs from all tropical populations of *Caulerpa* in lacking ITS polymorphism, a fact that can be related to a prolonged confinement under aquarium conditions.

This finding has been confirmed in a repetitive analysis performed by Jousson et al. ([2.33] 2000), that included strains extracted from the Californian site. Eleven out of 12 Californian sequences were found to be identical to all aquarium and most Mediterranean sequences (fig. 2.h-right). 64 sequences from the Mediterranean-aquaria-California isolates were identical, whereas five were slightly divergent (from 0.4 to 1.1%). The sequences reveal a relatively robust clade (80% bootstrap value) grouping Californian, Mediterranean, aquaria and some Australian sequences together. Thus, confirming that even the Californian species belong to the aquarium strain.

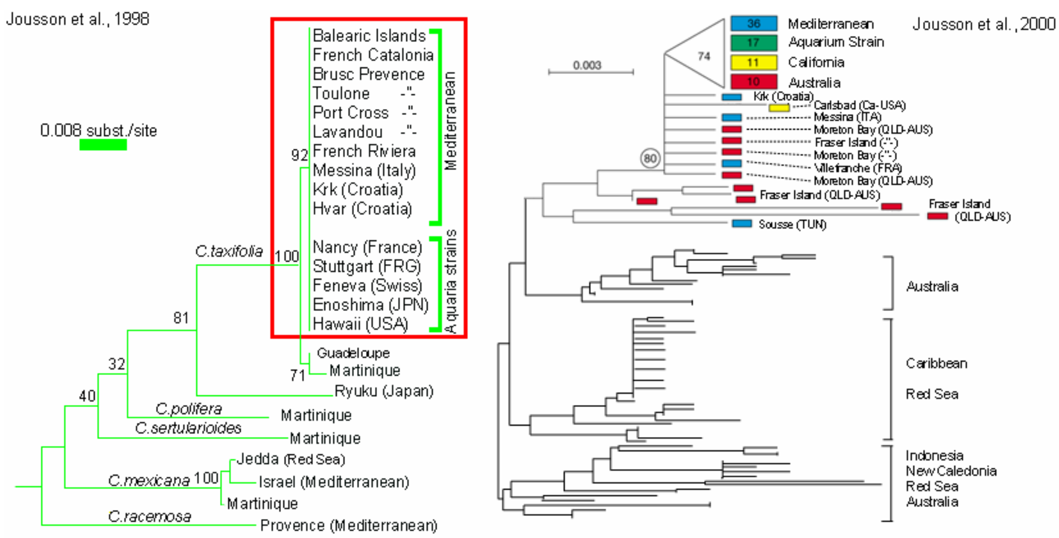


Fig.2.h: Phylogenetic analysis of *C.taxifolia*.

Benzie et al., ([2.34]2000) tested the genetic relationships of the fine and robust morphological types of *C.taxifolia* and assessed the extent of genetic variation among widely separated populations of aquarium *C.taxifolia* with similar morphology of those found in Australia. Furthermore it was tested if *C.taxifolia* could be an ecomorph of *C.mexicana*. The findings revealed that allele frequencies were markedly different between *C.mexicana* and *C.taxifolia* populations. A cluster analysis grouped all of the *C.mexicana* populations together and separately from all of the *C.taxifolia* populations (fig. 2.i). The genetic distances between these species were generally far greater than between populations of each species.

Although populations of *C.taxifolia* that can develop a phenotype more akin to *C.mexicana* (Chisholm et al., 1995), both allozyme analysis and ribosomal sequence data confirm the true genetic differences among them ([2.35]Olsen et al., 1998) and that there are indeed two separate taxa, however some populations of *C.taxifolia* may overlap in morphology with *C.mexicana*.

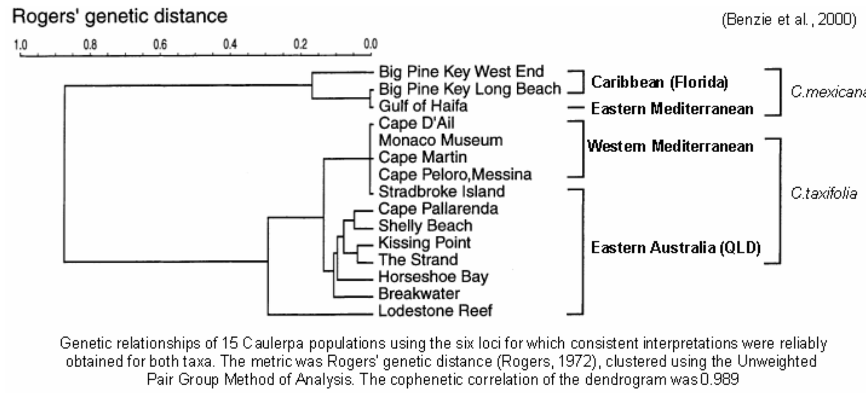


Fig.2.i: Phylogenetic analysis of *C.taxifolia*.

Wiedenmann et al., ([2.30]2001) compared samples from 11 locations in the Mediterranean Sea with 3 representatives from public aquaria. The uniformity of hybridization patterns indicates that representative specimens from the Mediterranean and aquaria belong to the same clone. The slight differences in hybridization patterns in *C.taxifolia* from *Manly Harbour* (Australia) suggest that it carries very similar chloroplast and mitochondria traits. The Australian population of *Manly Harbour/Moreton Bay* is well suited for comparative studies of the role of *C.taxifolia* in a non-Mediterranean ecosystem because of the close relationship to the aquarium strain.

The comparative results of *C.taxifolia* strains from aquaria, the Mediterranean Sea and from *Manly Harbour* (Australia) are shown in fig.2.j in which $(CAC)_5$-hybridised Southern blots of total DNA after TaqI digestion (restriction patterns in ethidium-bromide stained agarose gels) have been performed.

In contrast, the control sample (left lanes 1-2) clearly distinguishes them from the aquarium strain (*C.prolifera, C.taxifolia* from *Martinique*). The aquaria strains from *Stuttgart* and *Enoshima* reveal identical restriction patterns as samples from the Mediterranean Sea (*Monaco, Krk, Sicily, Mallorca, Elba* 1-3, *St.Cyprien* 1-3). Only slight differences in the position of a single band (indicated by the circle) were detected between the sample from *Manly Harbour* (lane 16) and the aquaria specimens.

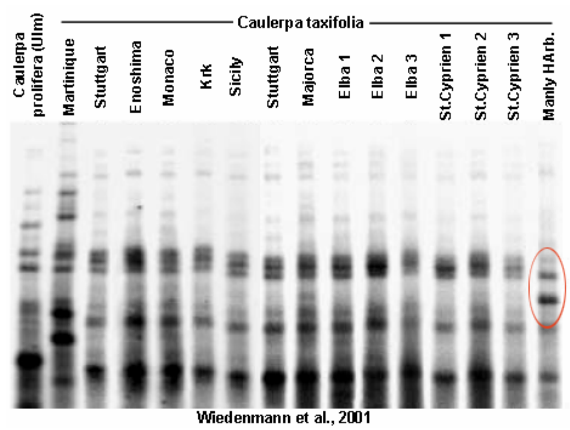


Fig.2.j: Fingerprints of *C.taxifolia*.

3. Invasive Properties, Reproduction & Nutrient Dynamics of *C.taxifolia*

3.a. Invasive Properties: An invasive organism is defined as a species that fulfills all of the following criteria (modified after [3.1]Carlton, 1985; [3.2]Ribera & Boudouresque, 1995; [3.3]Williamson & Fitter, 1996; [3.4]Boudouresque & Verlaque, 2002).

(i) it colonizes a new area where it was not previously present;
(ii) the extension of its range is linked directly or indirectly to human activity;
(iii) there is a geographical or genetic discontinuity between its native area and the new area (remote dispersal or genetic modification). This means that the occasional advance of a species at the frontiers of its native range (marginal dispersal) is not taken into consideration;
(iv) finally, new generations of the allochthonous species are born in-situ without human assistance, thus constituting self-sustaining populations: the species is established, i.e. naturalized.

The two additional questions which warrant mentioning are:

(i) why do some species become invasive and others not? and also,
(i) is it possible to predict the invasiveness of an introduced species?

Amongst invasive fresh water plants, several common features, likely to account for their success, have been identified ([3.5]Ashton & Mitchell, 1989; [3.6]Pieterse, 1990):

(i) vegetative reproduction is usually the commonest, and often the only method of reproduction; as a result, one viable propagule is sufficient to start a new colony;
(ii) vegetative reproduction is prolific;
(iii) habitat requirements are flexible;
(iv) they tolerate environmental fluctuations and extremes of the invaded habitat;
(v) there is a similarity between the native and recipient habitat;
(vi) they are free from predators and diseases characteristic of their native range.

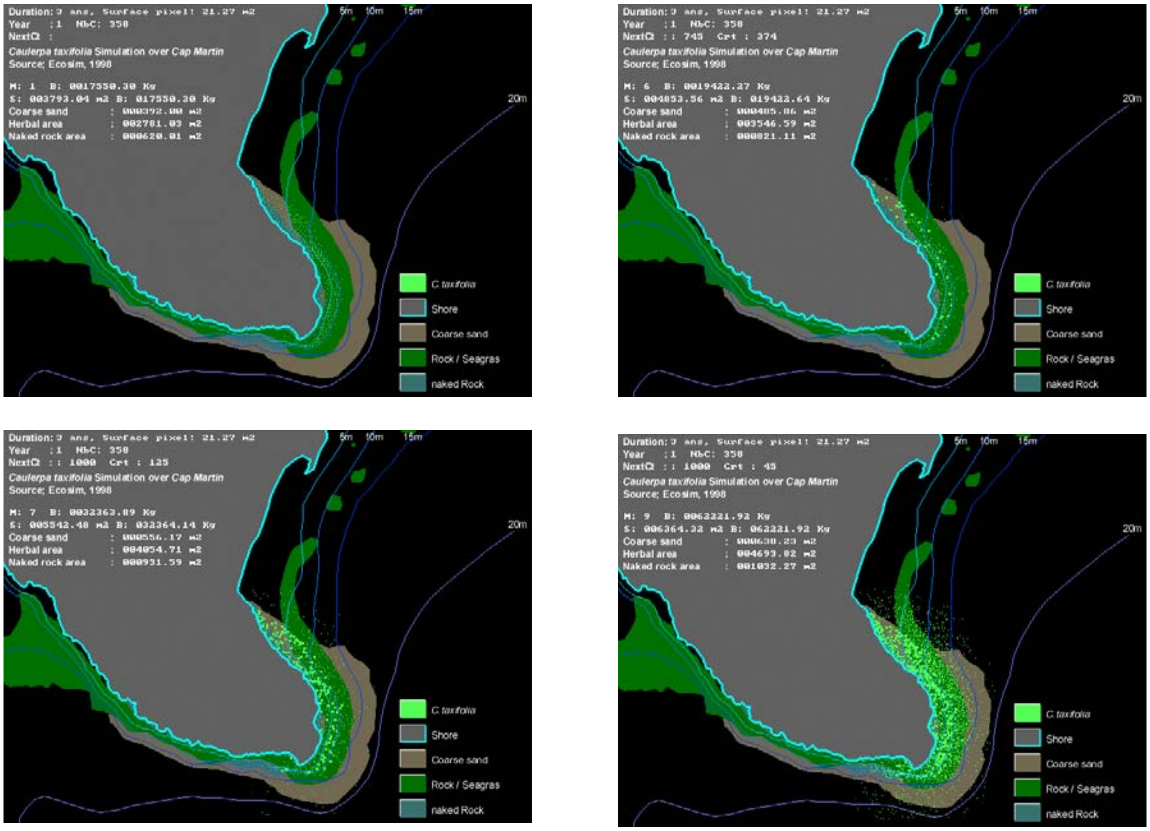
At any given site, this ubiquitous alga is able to colonize most if not all habitats. In the Mediterranean it has invaded *Posidonia* and *Cymodocea* seagrass beds, rocks with photophilic or sciaphilic alga, steep cliffs with sponges and sea fans, coarse and muddy sands, with a coverage of up to 100% maintained throughout the year ([3.7]Vaugelas et al., 1999). Today, more than 90% of the total cover of *C.taxifolia* is found in the north-western Mediterranean ([3.8]Meinesz et al. 2001). During the first decade of the Mediterranean invasion, the alga's distribution pattern was rather more irregular and concentrated to zones where heavy development took place or in the vicinity of poorly treated wastewater outlets. The discharge of huge amounts of raw sewage took place till 1981/82, and even as late as 1990, the main sewer outlet for Monaco discharged primary treated wastewater only 400m offshore. This remark is important as wastewater contains large quantities of phosphorous that in turn exhibit extremely low capacities to transform NH_4^+ into NO_2^- and NO_3^- - a fact that can still be found in the geo-chemical signature of the sediments ([3.9]Chisholm et al., 1997).

With the regression of the once abundant autochthonous flora (*P.oceanica* meadows, [3.10]Olivier, 1929) that started some 30 years before the invasion of *C.taxifolia* took off, it was an ideal hub for *C.taxifolia*'s to invade the N-W Mediterranean ([3.11]Meinesz and Laurent 1978; [3.12]Falconetti and Meinesz 1989; [3.13]Chisholm et al., 1997).

Fig.3. Underwater sewage outlet.

Analysis in that area have revealed that total organic material, i.e. including size fractions of decayed seagrass leaves, rhizomes and raciness of *P.oceanica* (>2mm), was high in all sediments providing excellent conditions for *C.taxifolia* growth. Concentrations of non-crystalline sulfide were high in all potential locations.

With the Ligurian Current flowing from east to west along the north Mediterranean coast and along with it contaminants traveling huge distances from their site of origin, it is therefore not coincidental that *C.taxifolia* has invariably begun proliferating around the periphery of headlands ([3.14]Meinesz et al. 1994), which are both zones of current impaction and favored sites for wastewater discharge ([3.9]Chisholm et al., 1997).



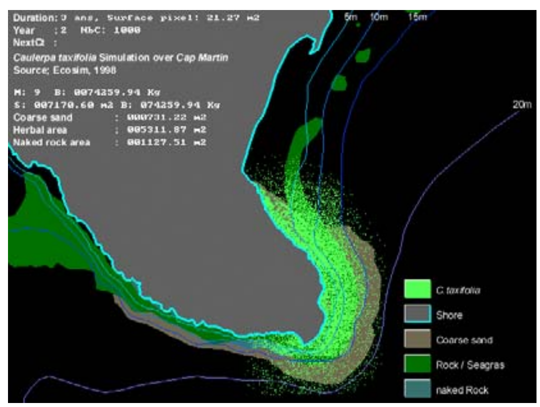


Fig.3.a: *C.taxifolia* EcoSim-Simulation 1998. Gradual increase in C.taxifolia coverage over a three-year period (from left to right).[3.6a]

Since the launch of the invasion in 1984, the alga's spread continued resulting in coverage indices in the most affected benthic areas of up to 100% between depths of 1 to 35m. Below this depth, it has been observed - though at much smaller densities - as far down as 100m ([3.15]Belsher & Meinesz, 1995). Such depths are unknown for the tropical strain of *C.taxifolia*: 30m at Papua-New Guinea, 32m at Tahiti, 50m at New Caledonia, 32m in the tropical Atlantic around Virgin Islands, *St. Croix* ([3.15]Belsher & Meinesz, 1995).
In addition, this aquarium strain of *C.taxifolia* can survive out of water and under humid conditions, for up to 10days ([3.16]Sant et al., 1994). It can florish in a wide variety of habitats, including sandy bottoms, rocky outcroppings, mud, sheltered bays, and natural meadows. *C.taxifolia* is able to withstand severe nutrient limitation ([3.17]Delgado et al., 1996). Furthermore, the algae adapts to any milieu, be it a polluted port or a clean, isolated bay.

Potential invasion sites are first colonized around headlands and were drifting algal fragments can attach. With its ability to form dense carpets, the aquarium strain is capable of extremely rapid growth resulting in exceptionally dense meadows. This is in sharp contrast to the tropical strain of *C.taxifolia* where it occurs in isolated and patchy aggregations ([3.18]Meinesz & Hesse, 1991).

So far the aquarium strain has not been found, where water temperatures fall below 20°C. Yet, it has been shown that the alien version of this plant can survive in the laboratory at temperatures of 10°C for three months (its lethal threshold value is at 7°C). In the Mediterranean, where winter temperatures generally drop to just 13°C these algae maintains its dense underwater meadows - despite signs of chlorosis ([3.19]Gnassia-Barelli et al., 1995).

During summer (June to September) the thallus of the aquarium alga attains extreme growth rates of up to 32mm of new stolon per day and a new frond every other day (month of August) resulting in frond densities of approximately 5000 fronds/m^2 ([3.20]Meinesz, 1995). An extremely densely covered single square meter of seafloor can be matted with 230m of stolons, from which emerge up to 8000 leafy fronds ([3.18]Meinesz & Hesse, 1991).

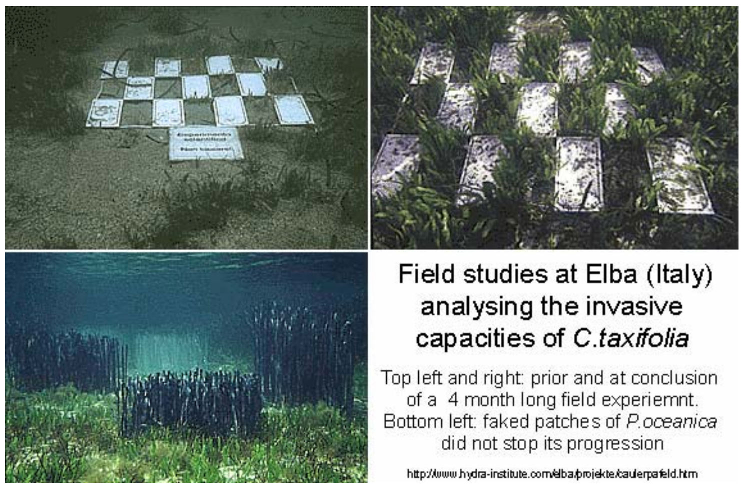


Fig.3.b: Invasive properties of *C.taxifolia*.

Although large-scale die-off occurs in shallow water (0.5–2 m) in most waterways during the cooler months and particularly after heavy rainfall (compare fig.1.f - Chapter-1, [3.20a]Creese et al., 2004), this die-back as a result of decreased temperatures, lower salinity and increased turbidity, is only temporary as the algae picks of again during the warmer months to make up lost ground and to proliferate beyond pre-established limits. Thus, the rate of expansion of this invading species as well as the impacts noted upon the environment, along with the feature of asexual reproduction that will be discussed shortly, assigns this weedy species a catastrophic and property represents a major risk for shallow underwater ecosystems of the Mediterranean.

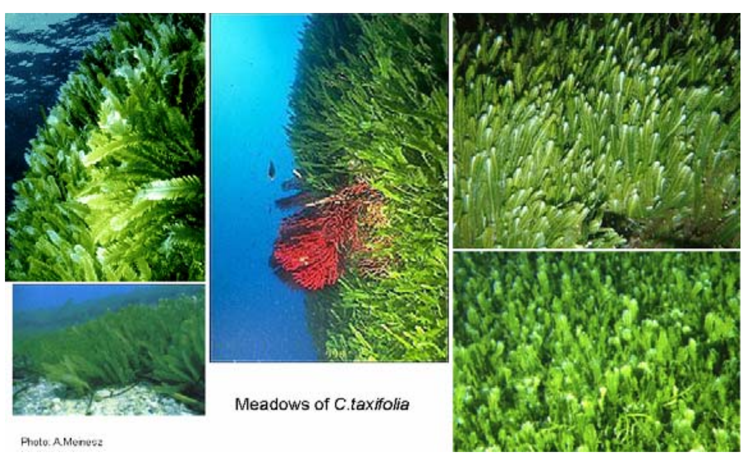


Fig.3.c: Mediterranean *C.taxifolia* meadows.

3.b. Reproduction: In the aquarium strain of *C.taxifolia* only male gametes have been observed and this strain is apparently only vegetatively spreading ([3.21]Zuljevic and Antolic, 2000). Sexual reproduction of this aquarium strain appears to be a stochastic event (Clifton and Clifton, 1999) as it has been only observed in temperatures above 25°C ([3.21]Zuljevic and Antolic, 2000).

Caulerpa's life cycle is therefore poorly understood. But when it does occur, it involves "holocarpy", in which the entire protoplasm gives rise to biflagellate gametes at once; i.e. almost all of the protoplast of a thallus is converted into gametangia containing the biflagellate dissimilar (anisogamy) motile gametes, which are discharged through papillae. Upon merger of the gametes, the zygote develops into a protonema, which then forms a typical diploid (2n) thallus ([3.23]Silva, 2002). Most siphonous green algae are primarily diploid; the gametes are the only haploid cells in the life cycle ([3.24]Raven et al., 1992). Since *C.taxifolia*'s life cycle is poorly understood, it could well be that the mature sporophyte releasing quadri-flagellated zoospores that in turn would give rise to the gametophyte (n), is inexistent (see fig. 3.d).

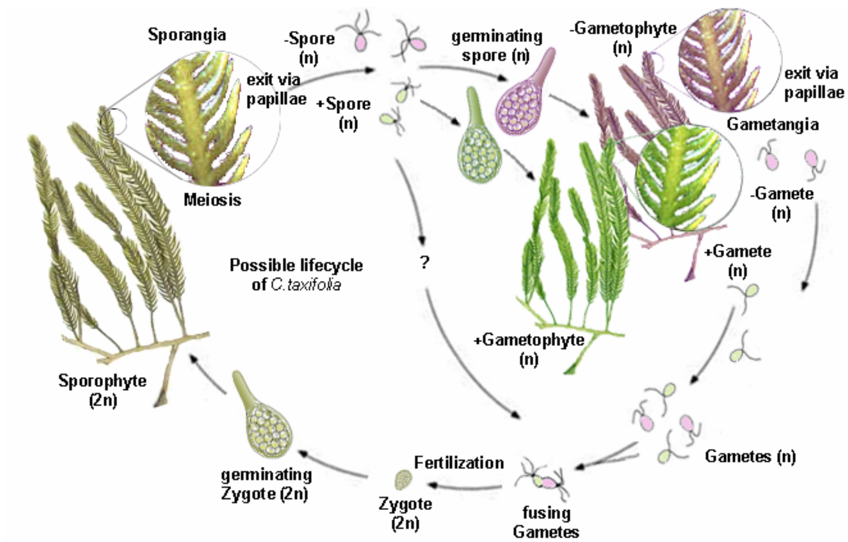


Fig.3.d: Lifecycle of *C.taxifolia*.

According to Meinesz, *C.taxifolia* is able to disperse a shower of male and female gametes that pair up and fuse to form a zygote (new plant) under lab-conditions. But over more than 10 years that he has observed this species in the Mediterranean and in the lab, he has never seen evidence of sexual reproduction. In the wild, though, the only reproductive cells released are male (fig.3.e), confirming existing evidence that all *C.taxifolia* in the Mediterranean are clones of that single aquarium plant release in 1984.

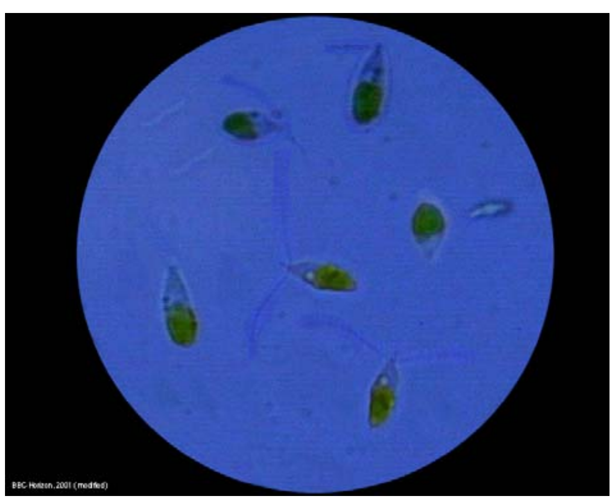


Fig.3.e: Gametes of *C.taxifolia*.

Fig.3.f: Vegetative reproduction of *C.taxifolia*.

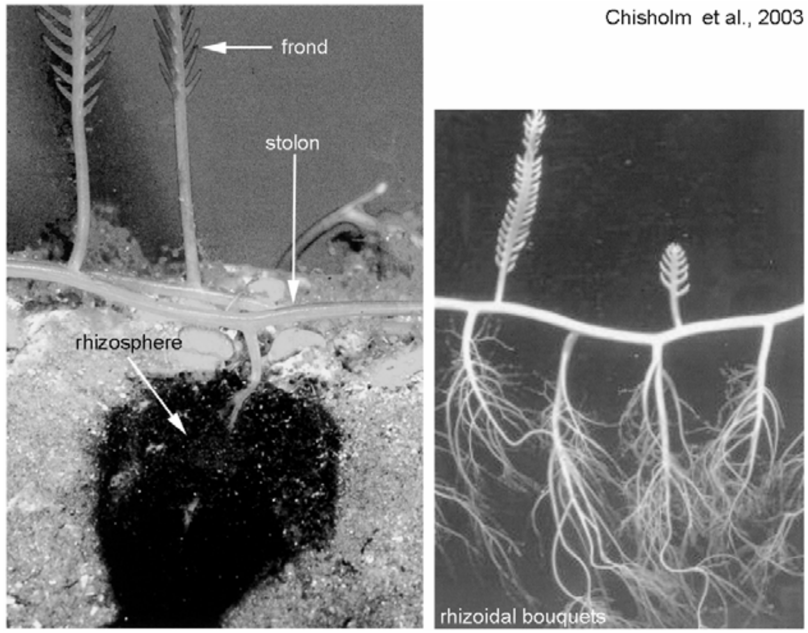
Genetically, this invasive species shows relatively little variation, thus vegetative reproduction by fragmentation is the most common mode of proliferation (asexual or clonal propagation). The break-up of thalli (mediated via anchor damage, fishing gear or storm activity as small as $1cm^2$) gives rise to new colonies that usually appear in 2 to 10m deep water ([3.25]Meinesz et al., 1993) during summer and fall when growth rates are highest (compare fig.5.f of Chapter 5).

3.c. Nutrient dynamics: Little is known about the nutrient dynamics of *C.taxifolia* such as its preferred high affinity for nutrients, its ability to take up nutrients from the sediments via rhizoids, and its capacity to store nutrients. Studies on other members of Caulerpales suggest the existence of some of these features. *C.taxifolia* can utilize nutrients and carbon sources from the sediment via uptake through the rhizoids and associated bacteria (Chisholm et al., 1996), even in eutrophicated, anoxic sediments ([3.26]Chisholm and Jaubert, 1997). Therefore the alga was shown to be tolerant to shading conditions ([3.27]Komatsu et al., 1997) enabling growth in areas where photosynthesis is light-limited as a result of greater depths or during the darker winter months.

As mentioned above, enrichment of sediment with inorganic components originating from wastewater discharges, are some of the prerequisites for the alga's success in the N-W Mediterranean. This assumption was confirmed by recent studies indicating that *C.taxifolia* only succeeds in colonization sparse or patchy *P.oceanica* communities ([3.28]deVillele & Verlaque, 1995). Once *C.taxifolia* proliferates due to the heavily enriched inorganic material perfused with hydrogen sulfide, it alters the sediment's "microclimate" into anoxic conditions (see fig.3.g). These are usually conditions that kill most autochthonous algal species that have been tested in studies on algal mats in eutrophicated waters ([3.29]Schramm & Both, 1981). When there is no growth of fronds (winter) and photosynthesis is weak (>30m), copper, iron, manganese, and lead concentrations were high, indicating an adsorption process from the substrate for uptake ([3.30]Gnassia-Barelli et al., 1995).

Field and laboratory observations have shown that sediments containing decaying organic matter of out-competed sea grass turn black in color once penetrated by the rhizoids of *C.taxifolia* (fig.3.g). The change in coloration is the result of redox conditions (bacterial reduction of sulfate to sulfide) favoring N_2-fixation, thus enhancing nutrient supply to the alga's rhizoids. In addition, *C.taxifolia* appears to activate the fermenting bacterial community by releasing photosynthetic product from its rhizoids in the form of simple sugars into the rhizosphere - similar to salt marsh and sea grasses. A process that can be simulated by simply injecting solutions of glucose or sucrose into unvegetated sediments ([3.13]Chisholm et al., 2003). *C.taxifolia*'s success thus stems from its ability to produce organic and inorganic nutrients from sediments via endocellular bacteria that potentially facilitate removal of toxic sulphide ([3.31]Chisholm et al., 1996).

Fig.3.g: Rhizosphere and rhizomes of *C.taxifolia*.



Chisholm's data indicate that the growth of *C.taxifolia* substantially enhances nitrogen production in high C:N sediments on dead *Posidonia* sediments in relatively low-nutrient seawater, thus facilitating organic matter turnover. *C.taxifolia*'s ability to stimulate nutrient turnover in substrata and then take up a proportion of the resulting nutrients via its subterranean rhizoids likely assists the remediation of sediments that are burdened by large quantities of refractory organic waste ([3.32]Chisholm & Moulin, 2003).

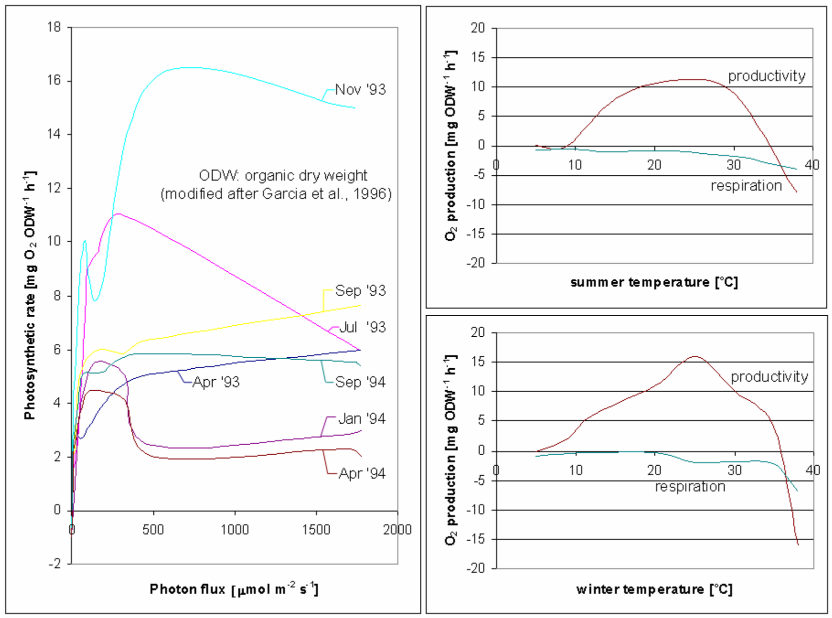
The same authors though predict that heterotrophic bacteria are the primary beneficiaries of N_2 fixation, thereby re-mineralizing C, N, and P. Once the transformation of the sediment's eutrophicated organic matter approaches completion, the abundance of *C.taxifolia* should decrease dramatically in the absence of an exogenous N supply of similar magnitude. This might explain some of the many boom-bust cycles of *Caulerpa* growth that have occurred in different environments ([3.33]LaPointe et al. 1994; [3.34]Panayotidis & Montesanto, 1994; [3.35]Doumenge 1995; [3.36]Davis et al., 1997). As the spread of *Caulerpa* might reach a plateau - be it by nutrient shortage or natural regulation (via a yet unknown predator - compare Chapter 5) the pollution prevention of marine habitats with wastewater, are just a few of a series of steps necessary to halt its hunger for new territories.

At 10m depth, the persistently high biomass of *C.taxifolia* in dense meadows of up to 613g/m^2 dry-weight (according to Verlaque & Fritayre (1994), which corresponds to a fresh-weight biomass of approx. 4kg/m^2 or more) represent an important nutrient trap, holding about 1g phosphorous/m^2 and 14g nitrogen/m^2. During summer and autumn when fronds of this alga reach their maximum length of 60-80cm, the biomass of *C.taxifolia* can exceed 10kg wet weight/m^2 ([3.18]Meinesz and Hesse, 1991; [3.38]DeVilléle & Verlaque, 1995).

These data are two-fold higher than the phosphorous contained in a seagrass bed of *P.oceanica*, though frequently lower than the amount estimated for populations of sublittoral algae ([3.20]Meinesz et al., 1995). In this regard, the siphonaceous nature of the thallus of *C.taxifolia* facilitates the nutrient translocation of the stored nutrients, as well as the reallocation of nutrients taken up from either the water column or the sediment. Likewise, nutrient recycling inside *C.taxifolia* meadows may have a pre-enrichment role in nutrient availability to the plant through nutrient reabsorption from dead and decomposing parts excreted by resident animals and bacterial activities.

In response to changes in light, temperature and nutrients, most of the Mediterranean seaweeds and sea-grasses have distinct annual cycles of biomass accretion and productivity. In fact this alga has a very low light compensation point and can grow in low light levels ([3.39]Garcia et al., 1996; [3.26]Komatsu et al., 1997). Most of the dominant seaweeds and sea-grasses in the Mediterranean display their lowest biomass gain and productivities during fall ([3.40]Ott, 1980, [3.41]Bay 1984, [3.42, 43]Ballesteros, 1989 & 1992). In contrast *C.taxifolia* displays the highest photosynthesis activity in November, coupled with large thalli dimensions ([3.28]Villele & Verlaque, 1995). Since *C.taxifolia* is a pseudo-perennial the high photosynthetic capacity during fall provides a competitive advantage to *C.taxifolia* over other species. Fig.3.h shows the short-term effects of temperature on light saturated photosynthesis (net productivity) and dark respiration rates of *C.taxifolia* collected in summer July '93 and winter Jannuary'94 (left). The right half shows the photosynthetic rates as a function of photon flux density for plants of organic dry weight of *C.taxifolia* collected at various times of the year from 9m depth at *Cap Martin*, France ([3.39]Garcia et al., 1996).

Fig.3.h: Effects of light and temperature on growth rate of *C.taxifolia*.



Although this species is pseudo-perennial ([3.20]Meinesz et al., 1995), the aquarium strain of *C.taxifolia* can resist hard winters showing vigority not present in its tropical counterpart. The aquarium hybrid prefers cooler water (around 10°C); its lethal minimum temperature in the Mediterranean is 7°C, lethal minimum temperature elsewhere is 14°C ([3.27]Komatsu et al., 1997); optimum growth temperature is 20-30°C; its lethal maximum temperature is 32°C. The year-round biomass accumulation of *C.taxifolia* has been estimated to average 5.5kg/m^2 (fresh-weight at Cap Martin, [3.20]Meinesz et al. 1995). Such production translates to a dry weight increase of 7.03g·m^{-2}·d^{-1} using a mean organic C to total dry weight ratio of 1.757:1 ([3.39]Garcia et al., 1996). In addition, sediment nutrient enrichment has been shown to increase growth of Mediterranean *C.taxifolia* ([3.44]Ceccherelli & Cinelli, 1997). Together all these features explain its outstanding growth ability and out-competing properties towards autochthonous species.

4. Effects on autochthonous species and C.taxifolia's toxicity

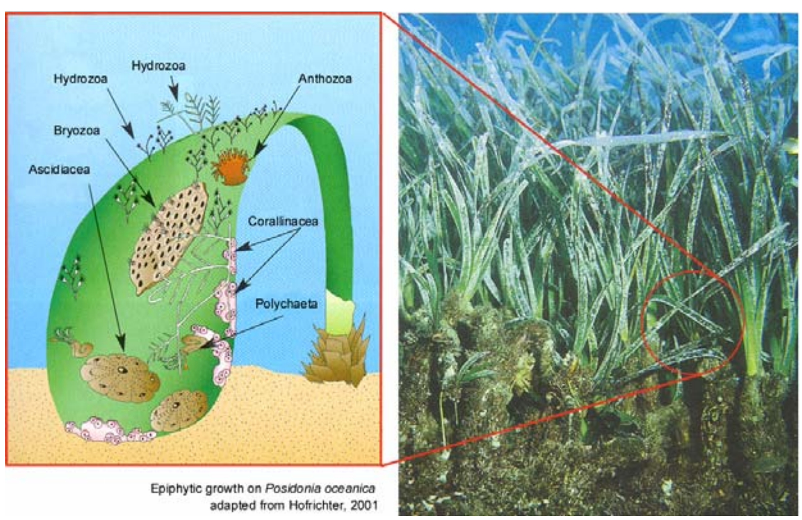
As *C.taxifolia* colonizes all types of substrata from the eulittoral down to 100m depth, inducing a homogenization of microhabitats and a reduction of the architectural complexity of the substratum ([4.1]Harmelin, 1996; [4.2]Harmelin-Vivien et al., 1999), a decrease in diversity and abundance of motile invertebrates was observed in the *C.taxifolia* meadows ([4.3]Bellan-Santini et al., 1996), as well as a persistent decrease in mean species richness, density and biomass of fish assemblages ([4.4]Francour et al., 1995; [4.2]Harmelin-Vivien et al., 1999), when compared to indigenous communities living in *P.oceanica* beds or in rocky areas. Relini et al., [4.5](2000) described qualitative and quantitative changes in fish communities during the replacement of the sea grass *Cymodocea nodosa* with that of *C.taxifolia* (at *Imperia*, western Ligurian Sea, Italy). In addition to the high fishing pressure (local/coastal fishing industry) and weak rugosity, they strongly impacted upon density and demographic structure of fish assemblages and ultimately leading also to a decrease of the abundance of larger-sized fishes.

As will be outlined in this part, the possible transfer of toxins through the food chain presents a toxicological risk not only for marine organisms exposed to it but also for man; i.e. certain mollusks feeding on *Caulerpa* showed a two- to three-fold concentration of toxic metabolites and became themselves toxic to predators, while human food poisoning resulting from the consumption of the Mediterranean bream *Sarpa salpa* has been already observed ([4.6]Spanier et al., 1989, [4.7a]Whitehead et al., 1986).

4.a. Effects on microbial loop: Soft sediment bottoms are not only characterized by a high infauna and epibenthic fauna, but are directly coupled to the microbial association of the sandy substrate. Particulate organic material and plankton organisms in the surface water are trapped and accumulate temporarily in shallow soft bottom sediment. Usually some of this carbon is available for consumption by benthic microbes in the system. However, the presence of an algal mat on otherwise unvegetated shallow soft bottoms considerably changes its ecosystem structure and functions. Rather than enabling a healthy microbial association to perform the nutrient conversion and especially under eutrophicated conditions, *C.taxifolia* takes over the "recycling activity". The altered nutrient dynamics of the sediment becomes evident in the net accumulation of organic matter. In the long term however, the organic enrichment leads to higher oxygen consumption that, together with the reduced water exchange, will result in decreased oxygen levels in both the water column and the sediment. The reduced oxygenation of the sediment causes the redoxcline to move towards the sediment surface further reducing the mineralization-capabilities of the microbial loop ([4.7b]Troell et al., 2004).

4.b. Effects on flora: Phytobenthic studies of highly infested areas around *Cap Martin* (Southern France) showed a drastic impoverishment of autochthonous communities. The creeping and erect axes of *C.taxifolia* shade off the light while its rhizoids trap and chemically alter the sediment. Most autochthonous algae tend to disappear quickly while crustose algae seem to be eliminated latest. The paucispecific *C.taxifolia* meadows tend to substitute for all sheltered algal infralittoral phytocoenosis which stands for a dramatic fall of richness and diversity of the littoral ecosystem.
Among the dozens of plants that are found in an intact Mediterranean ecosystem are the marine phanerogams of *Posidonia*, *Cymodocea* or *Zostera*.
Posidonia oceanica is a dark, gray/green endemic sea grass covering large areas of the seabed at depths between 30-40m ([4.8]Boudouresque & Meinesz, 1982), thus a fundamental key sea grass for that ecosystem. *Posidonia* meadows, like the other species of sea grass, not only bolster and protect the coastline, but it is one of the most important coastal primary producers ([4.9]Ott, 1980), act as a refuge, habitat, substrate for epiphytes, providing food and shelter for a huge variety of fish and invertebrates, and is the spawning ground and nursery for a countless number of species ([4.10]Meinesz, 2000 - see epiphyteic growth, fig.4).

Fig.4: Epiphytic growth on *P.oceanica* ([4.17a]Hofrichter, 2001).



Although *P.oceanica* possesses phenolic compounds that play an important role in the protection of these plants against competitors, predators and pathogens, that increase in stress-situations ([4.11]Agostini et al., 1998), it does not seem to effectively deter *C.taxifolia* from invading areas dominated by the seagrass *P.oceanica*; thereby killing up to 45% of *Posidonia* shoots in one year ([4.12]Villele & Verlaque, 1994). Similarly, other native seaweeds (e.g. *Cymodocea nodosa*) are being more or less totally replaced ([4.5]Relini et al., 2000; [4.13]Ceccherelli & Cinelli, 1997).

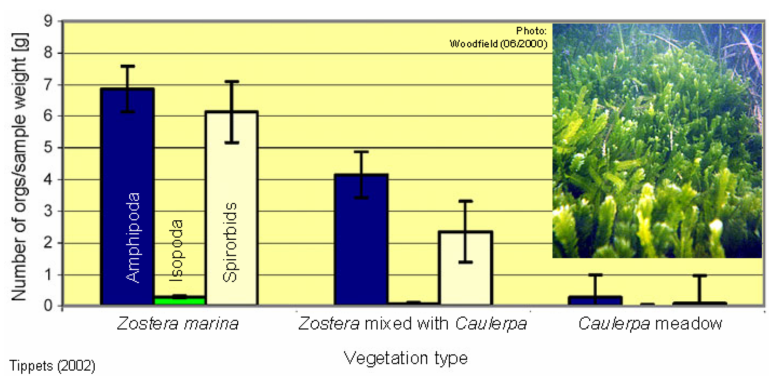
Since the phases of vegetative growth in *P.oceanica* and *C.taxifolia* are reversed ([4.14]Boudouresque et al., 1995; [4.15]DeVillele and Verlaque, 1995) the leaves of *P.oceanica* are larger in winter and spring than the fronds of *C.taxifolia*, which corresponds to the period during which growth of the latter species is at a minimum ([4.16, 4.17]Meinesz et al., 1993, 1995).

Both live and dead rhizomes of *P.oceanica* have in fact proved to be excellent substrate for the settling and development of this invasive chlorophyte ([4.18]Cuny et al., 1995). Consequently the patchy leftovers of *P.oceanica* along the southern coast of France could not cope with these altered unfavorable abiotic conditions and regressed giving way to this invasive alga in shallow waters and leaving behind decaying plant tissues. Indeed, the scale of destruction of the *Posidonia* meadows, which are irreversible, is alarming. Based on those suggestion Cecherelli & Cinelli, ([4.19]1999) predict that patches of *P.oceanica* at 10m depths are more vulnerable to algal invasions than at 2m depths. As *P.oceanica* leaves are moved by wave surge, they may sweep *C.taxifolia* from the substratum or abrade and tear it. If the negative effect of the whiplash is proportional to the length of seagrass leaves, it could explain the interaction of seagrass canopy height and density.

Fig.4.a: A running stolon of *C.taxifolia* (above) and *C.taxifolia* competing with *P.oceanica* (below)

During the Californian infestation focus on diversity of organisms in the three vegetation types was not the main aspect of the study, yet Tippets ([4.20]2002) was able to observe a decline in the richness between the native Californian eel grass *Zostera marina* and *Caulerpa* samples. A decline in certain "keystone" species, including amphipods, eventually results in a decline in all species dependent upon these organisms for food with ramifications extending up the food chain. As will be discussed later-on, one should not ignore the obvious presence in epiphytic end epizootic organisms commonly found on autochthonous seagrass species and as a result of the toxic cocktail of the allochthonous invader, the complete absence of such flora and fauna on its fronds. The effects of *C.taxifolia* invasions, and the ramifications on the benthic ecology of near shore lagoons, could be devastating to the basal levels of the food chain ([4.20]Tippets, 2002).

Fig.4.b: Effects on autochthonous fauna.



Due to the out-competing properties of *C.taxifolia* (eutrophicated sediments, growth rate, gigantism, etc. see above, it is becoming the canopy forming species in the Mediterranean ([4.17]Meinesz et al., 1995; [4.22]Ceccherelli and Cinelli, 1998); it occupies almost any substrate, and is therefore capable of replacing autochthonous floral competitors ([4.23]Verlaque and Fritayre, 1994; [4.24]Ferrer et al., 1997).

4.c. Effects on fauna (**invertebrates**): Belsher & Meinesz ([4.25]1995) observed that this invasive species suffocates numerous white Gorgonians *Eunicella verrucosa* at depths beyond 40m.
Bellan-Santini et al., ([4.26]1996) analyzed *C.taxifolia*'s effect on three important groups of invertebrates itself staple food for carnivorous fish: i.e. polychaetes, molluscs and amphipods are quantitatively and qualitatively dominant groups and at the same time exhibit varied types of trophic, reproductive, and dispersion strategies. Based on a low sampling effort, scientists have focused on results obtained by pooling the four seasonal samples at each location with the result that the numbers of individuals of mollusca, amphipoda and polychaeta in *C.taxifolia* meadows were greatly reduced.

Pedrotti & Lemée ([4.27]1999) could show that not only the marine microalga *Cricosphaera elongata* (phytoplankton) was subject to growth inhibition once incubated with organic extract of *C.taxifolia*. Even the herbivores feeding on it were affected. In particular a decreased survival rate of the sea urchin larva *Paracentrotus lividus* was found when the so exposed microalge was then offered to the larvae at different stages of development. Sensitivity to toxin-treated food depended on the larval stage at which exposure began. Larvae treated from first feeding (4-arm stage) with toxin-treated microalgae were most sensitive (25% survival, delay in development and metamorphosis of 32%). A *C.taxifolia*-diet begun at the 8-arm stage caused a decrease in survival and abnormal development; however, all the remaining larvae achieved metamorphosis (fig.4.c).

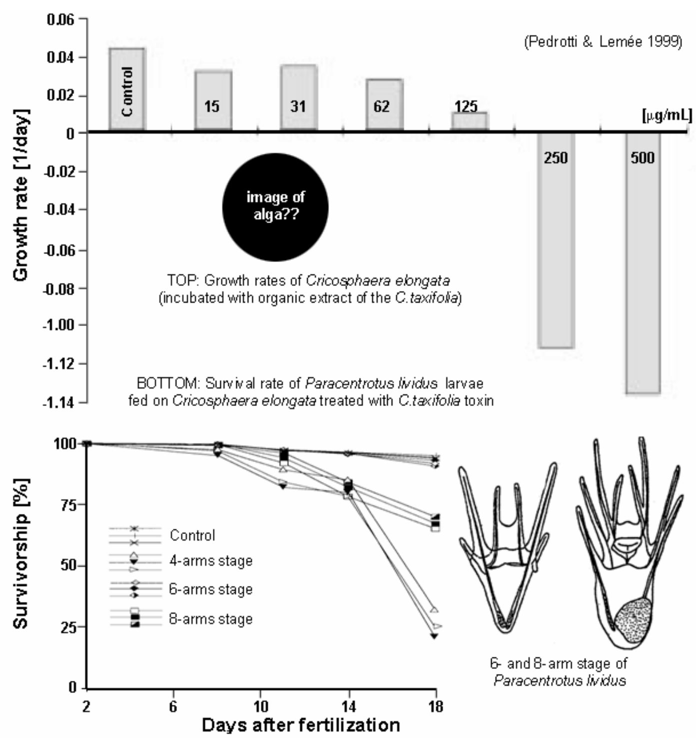
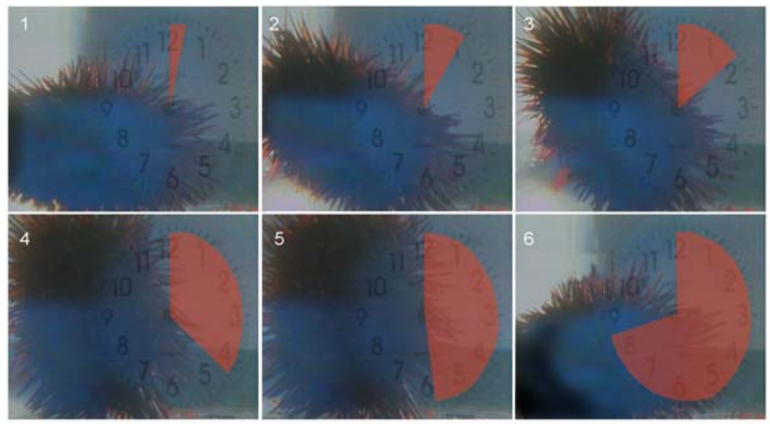


Fig.4.c: *Cricosphaera elongata* and *P.lividus*.

Boudouresque et al., ([4.28]1996) could show the toxic effects on adult *P.lividus*. They showed that urchins feed during the summer / autumn experiment were affected, five urchins died during the 2nd month in the *C.taxifolia* containing aquaria. By the 3rd month, all the urchins showed sub-lethal signs, including immobility, very high righting times and large areas bare of spines. It is already known that *C.taxifolia* has a repulsive effect against various herbivores, and particularly against the tropical Atlantic urchin *Lytechinus variegatus* ([4.29]McConnel at al., 1982); however, series of experiments involving one of the most important macrograzer confirmed the negative impact of *C.taxifolia* upon herbivores - with the fish *Sarpa salpa* being placed at the macroscopic end of the herbivorous spectrum ([4.30]Amade & Lemée, 1998). Although this effect was stronger in the summer-autumn period, than during the rest of the year, most of the urchins even preferred to eat their own waste or resorted to pieces of plastic, rather than touching the algae. Such behavior caused the urchins to die of hunger rather than consuming the available *C.taxifolia*. These findings were and still are very disturbing. It simply means that algal endotoxins disrupt the entire food chain and biodiversity of the affected ecosystem.

Fig.4.d: Turnover rates of adult *P.lividus*.



When placed up side down in a demonstration tank, *P.lividus* fed on native algae (e.g. *P.oceanica*) and in healthy conditions takes less than 1 minute to turn back over into its natural position.
A specimen exclusively fed on *C.taxifolia* requires 30min to achieve the same task (Photos: BBC-Horizon).

4.d. Effects on fauna (**fish communities**): The objectives of some studies done so far were also done to quantify the consequences of this expansion on fish assemblages. It was feared that the bright color pattern of *C.taxifolia* might change predation pressure of carnivores feeding on herbi- and carnivorous fish communities; in fact fish color pattern is believed to serve three main functions: thermoregulation, communication and antipredator adaptation ([4.31]Endler, 1978; [4.31]Crook, 1997) and are the results of selection pressures on the genetic structure of populations. Color patterns represent a good tool in understanding the adaptive mechanisms of a species to its environment ([4.30]Endler, 1978, 1980; Planes & Doherty, 1997).

Relini et al (2000[4.30a]), on the other hand, found that fish population changes in *C.taxifolia* meadows did not alter dramatically from those in *P.oceanica* meadows. Fish population still maintained the characteristics found in *C.nodosa* meadows and on sandy bottoms when *C.taxifolia* coverage was around 25%. Those authors concluded that the consequence of the allochthonous colonialization by *C.taxifioia* did not decrease fish biodiversity compared to autochthonous meadows. Bottom dwelling species (important commercial fish species) however suffered a severe blow due to the reduction of sandy seabed by the arrival of *C.taxifolia*.

In another study Francour et al., ([4.33]2002) focused on the color patterns of individuals of four Mediterranean labrid species, *Symphodus ocellatus, Symphodus roissali, Symphodus rostratus*, and *Coris julis*, living in dense *C.taxifolia* meadows. They were compared with those of the same species inhabiting their usual indigenous habitats, the *P.oceanica* seagrass beds and the shallow rocky areas. In *C.taxifolia* meadows the proportion of green morphs observed in *Symphodus ocellatus* and *Chromis julis* was significantly higher, particularly for small fishes. While *S.ocellatus* and *C.julis* settled in *C.taxifolia* meadows, *S.roissali* withdrew to shallow waters where *C.taxifolia* is not the dominant vegetation ([4.34]Francour et al., 1995).

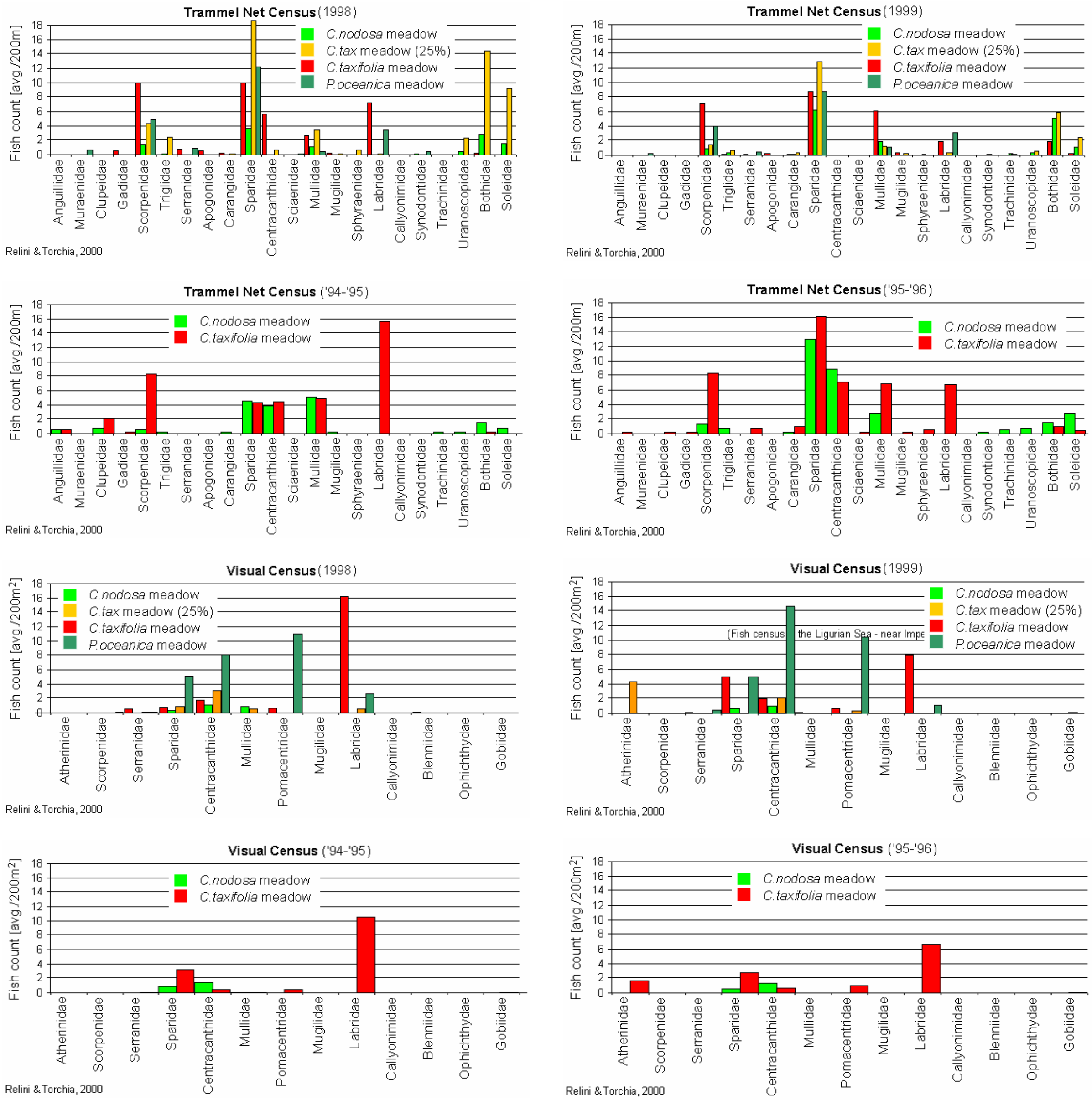


Fig.4.e: Changes in fish communities.

In its natural habitat, *S.rostratus* remained cryptic during the reproductive period, as it reproduced on the brown-dominated rocky area, whereas, in the new environment provided by the *C.taxifolia* meadows, reproductive individuals were more conspicuous to predators. Therefore, animals that rely on crypsis to avoid detection by predators commonly choose backgrounds upon which they will appear most cryptic ([4.35]Merilaita et al., 1999), by either moving away or trying to adapt via selection pressure - provided that they are able to survive on the altered food spectrum they rely upon.

4.e. Toxic agents in *C.taxifolia*: *C.taxifolia* is native to the tropics thus subject of intense herbivore activity leading to the development of efficient chemical defense and antifouling capabilities ([4.36]Paul and Fenical, 1986; [4.37]Faulkner, 1987; [4.38]Schwede et al., 1987). The cocktail of repellent toxins consists of **caulerpenyne** (CYN), oxytoxins, taxifolials and other terpenes (Paul, 2002). As CYN is the most predominant toxin, it is believed that toxicity is almost exclusively based on the acetylenic sesquiterpene caulerpenyne ([4.39]Paul, 2002) with a bis-enol acetate functional group. Caulerpin, on the other hand, is a hydrophobic macromolecule containing a cyclo-octatetraene ring pigment ([4.40]Ayyad & Badria, 1994). Both chemical structures are shown in fig.4.f. This molecule is synthesized in the fronds of the algae, thus concentrations are higher in the erect blades than in the rhizoids, where they are released into the surrounding sea-water or consumed by herbivores ([4.41]Pesando et al., 1996). CYN exhibits antibiotic activity ([4.36]Paul and Fenical, 1986, 1987; [4.42]Hodgson, 1984) and besides its cytotoxicity in mammalian cells including some eggs of some marine mammals ([4.43]Lemée et al., 1993; [4.44]Pesando et al., 1994), it is toxic for molluscs, sea urchins, herbivorous fish, and capable of killing off many microscopic organisms and other epiphytic organisms. CYN extract inhibits or delays the proliferation of several phytoplanktons of the marine food chain ([4.45]Lemée et al., 1997).

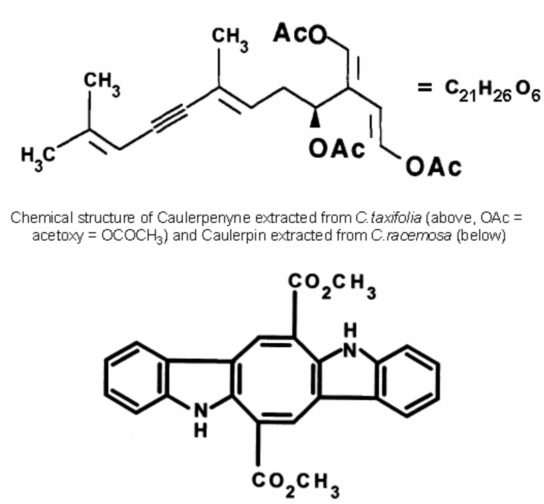


Fig.4.f: Structure of Caulerpenyne (CYN) and Caulerpin.

The aquaria strain of *C.taxifolia* contains higher CYN concentrations than the tropical strain ([4.46]Guerriero et al., 1992). *C.racemosa* as a Lessepsian migrant is likewise a tropical representative, but far less toxic than the *C.taxifolia*. The mean CYN values were found to be 80x higher in *C.taxifolia* than in *C.racemosa* ([4.47]Dumay et al., 2002) and are much higher than in other *Caulerpa* species ([4.48]Guerriero et al., 1994). CYN in the aquarium strain of *C.taxifolia* can account for up to 1.3% of the algal fresh weight or 2% or more of algal dry mass ([4.39]Paul, 2002).

4e. Toxicity of *C.taxifolia*: Under attack by herbivores - in the tropics by rabbit-fish (Siganidae) and surgeonfish (Acanthuridae; [4.39]Paul, 2002) - the liberation of chemical precursors convert CYN into a potent feeding deterrent ([4.49]Jung & Pohnert, 2001). Since CYN is reportedly stable in pure seawater, the rapid conversion of it in an almost neutral environment (pH 6-7) suggests the involvement of an enzymatic reaction. This wound-activated enzymatic transformation enables *C.taxifolia* to store the less reactive CYN that can be transformed to aggressive metabolites when under attack. Since the reactive aldehydes are present within seconds after tissue damage, they might act locally as defensive metabolites during the relatively slow feeding process by slugs or sea urchins.

Possible metabolites after esterase-action from a wounded *C.taxifolia* on CYN are shown in fig.4.g (uppermost line); e.g. acidic deacetylation of caulerpenyne in the presence of 2,4-dinitro-ohenyl-hydrazine. For clarity tetraenes resulting from H_2O elimination are omitted and only the most stable derivatisation product is shown ([4.49]Jung & Pohnert, 2001). Line 2, 3, and 4 of fig.4.g illustrate a few synthetic transformations from CYN isolated from *C.taxifolia*. However, no synthetic route toward CYN has been reported to date. Retrosynthesis scheme outlined the strategy for synthesizing CYN and one of its metabolites taxifolial A, which can be considered as a good precursor of CYN ([4.50]Parrain & Santelli, 2002).

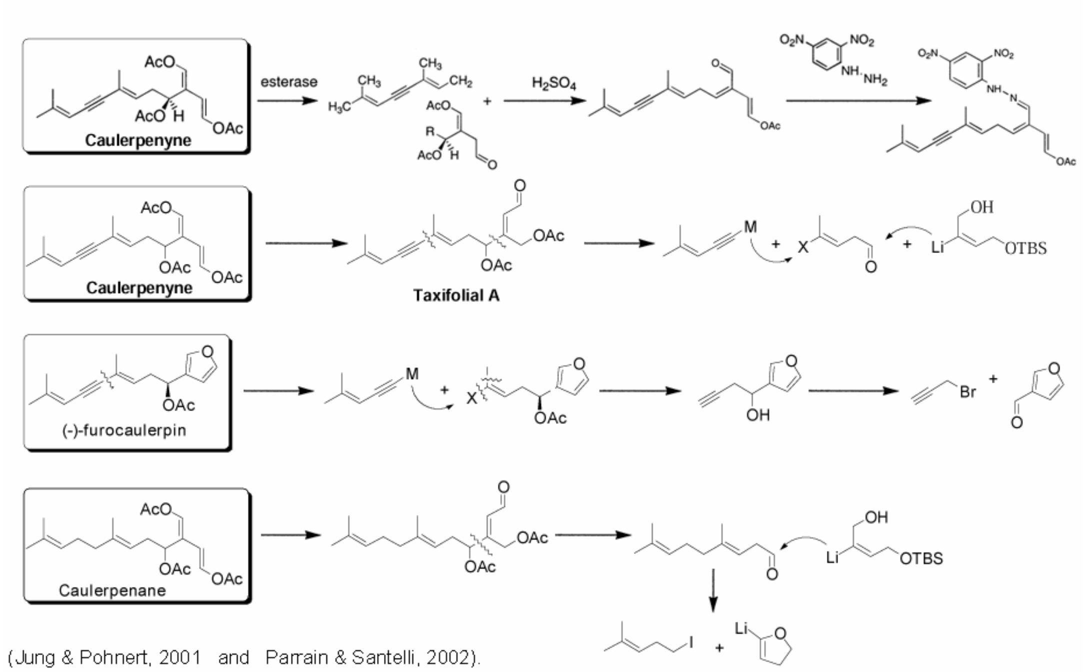


Fig.4.g: Caulerpenyne reactions

Caulerpa's toxicity is highly seasonal; the heaviest disturbance occurred in summer and autumn when the size of *C.taxifolia* and its terpenoids production peaked at a maximum. Epiphytic and epizootic growth on *C.taxifolia* is insignificant except in spring when endotoxin concentration is lowest ([4.51]Amade & Lemée, 1998). The annual decrease in CYN concentrations, with the concomitant increase in frond length, under conditions of competition with *P.oceanica* is a likely result of a modified metabolism in the alga as the increased energy allocated to growth of fronds would be at the cost of another function ([4.52]Dumay et al., 2002).

Usually during late winter, the cover of *C.taxifolia* generally decreases (with algal fronds much smaller, [4.17]Meinesz et al. 1995), only to pick up again during the warmer months to recuperate lost terrain. This periodic cycle correlates with its high growth rate, its total substrate occupation, light access, and sedimentation rates. This oscillation is reflected in the CYN concentration of the alga's frond wet weight: from to 0.2% in spring to 13% in summer ([4.53]Amade et al., 1994). The sketch at the top left in fig.4.h shows the concentration of CYN in seawater as a function of time and toxicity (left). Whereas the image at the bottom right of the same figure displays the amounts of CYN in fronds collected monthly at 5m depth in *Cap Martin* (France) in relation to the water temperature. On the other hand, the image at the bottom left reveals CYN concentrations as a function of the season and superimposed the inhibition of sea urchin egg cleavage in the methanolic extracts of *C.taxifolia* fronds (collected monthly at 5m depth at *Cap Martin*, France). This inhibitory effect is pictured in the top right part of figure 4.h. Even if CYN appears to be broken down in seawater, thus limiting its action to close-range vicinity of the algae, the degraded products are toxic and may influence the planktonic compartment as well as larval recruitments. In fact CYN is known for its repulsive and antifouling effects ([4.54]Meyer and Paul, 1992) confirming the presence of highest CYN at the surface of the alga with a negative gradient in the immediate seawater environment ([4.51]Amade & Lemée, 1998).

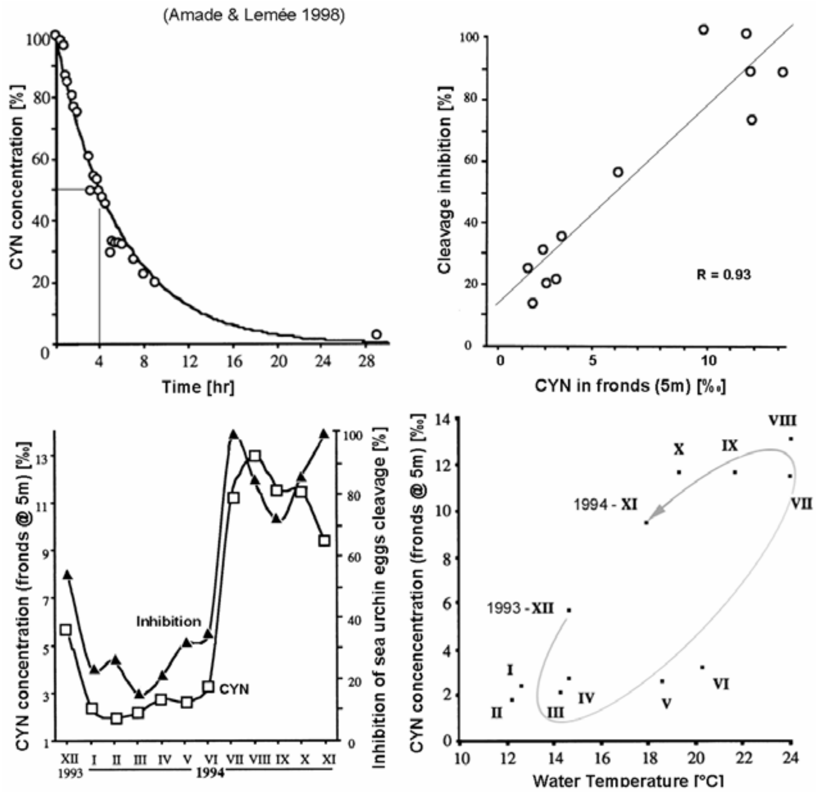


Fig.4.h: Caulerpenyne concentrations and *P.lividus* mortality.

C.taxifolia protects itself by producing substances that are toxic to the Mediterranean's two main macro-herbivores, sea urchins and their eggs (besides those of hamsters and mice tested in the lab) ([4.43]Lemée et al., 1993), and the herbivorous fish *Sarpa salpa*. Data collected provide evidence that CYN:

(1) exhibits antiproliferative activity in bacterial cultures of *Planococcus* ([4.55]Giannotti et al., 1994);
(2) toxic to ciliates ([4.56]Dini et al., 1994);
(3) inhibits first cleavage of sea urchin eggs without affecting fertilization by reducing cytosolic ATP-dependent Ca^{2+} accumulation;
(4) blocks the S-phase and mitotic cycle of sea urchin embryos at metaphase by modifying microtubule networks ([4.57]Barbier et al., 2001) and inhibiting the stimulation of nitrogen-activated protein kinase / phosphorylation ([4.41]Pesando et al., 1996);
(5) while Fischel et al., ([4.58]1995) and Barbier et al., ([4.57]2001) reported about the antiproliferative as well as growth-inhibitory effects of sesquiterpene in eight cancer cell lines of human origin (similar results have been obtained with in vitro tests of caulerpin by Ayyad & Badria ([4.40]1994));
(6) CYN is known to induces neurological disorders (i.e. amnesia, vertigo, and hallucinations, reported by [4.59]De Haro et al., 1993) on patients with food poisoning due to the ingestion of *Sarpa salpa* that fed on *C.taxifolia*);
(7) Brunelli et al., ([4.60]2000) and Barbier et al., ([4.57]2001) further found that CYN, besides its inhibitory effect on the Na^{+}/K^{+}-ATPase, also affects some other ion channels accounting for reduced after-hyperpolarization amplitudes and the decrease of cellular membrane resistance;
(8) Schröder et al., ([4.61]1998) have shown that multixenobiotic resistance (MXR) membrane pumps - present in marine organisms - are negatively affected by CYN and caulerpin; i.e. otherwise non-lethal concentrations of environmental toxins in combination with suppressed MXR mechanism resulted in a strong apoptotic response of target cells.

As species-poor meadows of C.taxifolia cover the infralittoral zone, it replaced the rich natural algal populations along with the disappearance of many of the normally occurring species associated with it, resulting in a drastic reduction in the richness and diversity of the Mediterranean littoral ecosystem. Due to the synthesis of toxic secondary metabolites (mono- and sesqui-terpenes) *C.taxifolia* has another advantage over the native seaweeds and sea-grasses. Eventually, the effect of the aquarium strain *C.taxifolia* in the Mediterranean is a major ecological event not only by protecting itself from predation ([4.14]Boudouresque et al., 1995), but also by significantly decreasing species diversity and fish biomass when compared with the *P.oceanica* beds. All this suggests that coastal ecosystems are clearly under threat, which will result in total uniformisation of underwater landscapes and their associated populations.

5. Control Measures, Strategies and Prevention

Various attempts that range from manual uprooting, mechanical means (underwater suction devices), physical control with dry ice, to chemical intervention utilizing household bleach (chlorine) and other chemicals have been tried to halt the spread of this invasive species. Accidentally, Meinesz and his colleagues stumbled over a more promising alternative, involving some selected predators that are able to feed on this particular mutant.

5.a Biocontrol via potential predators of *C.taxifolia*: Since 1994, the potential use of four ascoglossans (Mollusca: Opisthobranchia) as biological control agents against *C.taxifolia* (and *C.racemosa*) have been examined. These mollusks make incisions on all parts of *C.taxifolia*. They perforate the cell wall with its uniserial radula and sucks up a small portion of the algal contents, leaving light colored markings on the alga. Most ascoglossan sequester secondary metabolites from its diet for their own defense ([5.1]Paul & Hay, 1986; [5.2]Jensen, 1994) thus storing and using caulerpenyne (CYN) from *C.taxifolia* as a feeding deterrent.

Oxynoe olivacea is a Mediterranean ascoglossan species, which is scarce in meadows of their usual food (strictly stenophagous on *C.prolifera*). It has become an adapted feeder on the invading tropical alga *C.taxifolia* ([5.3]Thibaut & Meniesz, 2000). It has a partial shell to protect its reproductive organs and digestive glands. Unfortunately, its grazing pattern is correlated with water temperature. Feeding at and above 22°C the number of incisions is almost double that observed at 16°C (at and above 16°C it is still three times that observed at 13°C). A single specimen can destroy 0.7-1.6cm/day of the alga's frond (5cm in 3-7 days).

Fig.5.b: *Oxynoe olivacea*.

Its larval stage is planktothrophic, rendering a possible use as a biological control against *C.taxifolia* only possible through an artificial enhancement of its populations after cultivation of the veligers and release of juveniles during the winter season.

Oxynoe azuropunctata is also a shelled species feeding exclusively on Caulerpales (stenotrophic). This species has a higher feeding rate than either *O.olivacea* or *L.serradifalci*; an individual is able to destroy a 3-4.5cm of frond per day ([5.3a]Meniesz et al., 1996). Unfortunately, this species has a brevipelagic larval phase ([5.4]Clark et al., 1979; [5.5]Clark & Jensen, 1981), diluting the recruitment of progenic predation by this species.

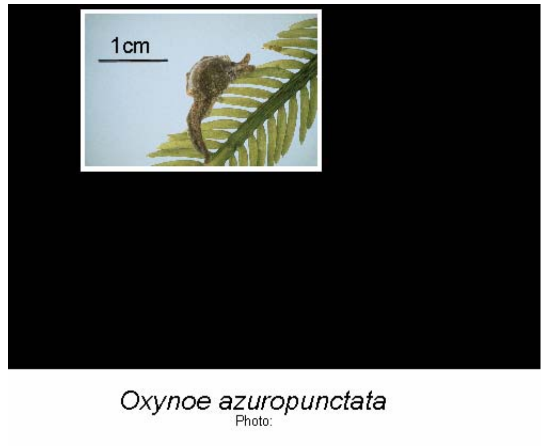


Fig.5.c: *Oxynoe azuropunctata*.

Lobiger serradifalci, another shelled species native to the Mediterranean that naturally feeds on *C.prolifera* has been observed to settle and feed on *C.taxifolia*. However, rather than helping to control the spread of this introduced algae it may in fact be hastening its spread. In the Mediterranean, at least, *C.taxifolia* is reproducing vegetative, by small pieces breaking off and growing into new plants. Unfortunately, *L.serradifalci* feeds by piercing holes in the cell wall, which weakens the plant and allows pieces to break off, so hastening the spread of the algae. Like *O.olivacea* and *O.azuropunctata* it has a planktothrophic larval stage; thus requires artificial enhancement of their populations after cultivation of the veligers and release of juveniles during the winter season.

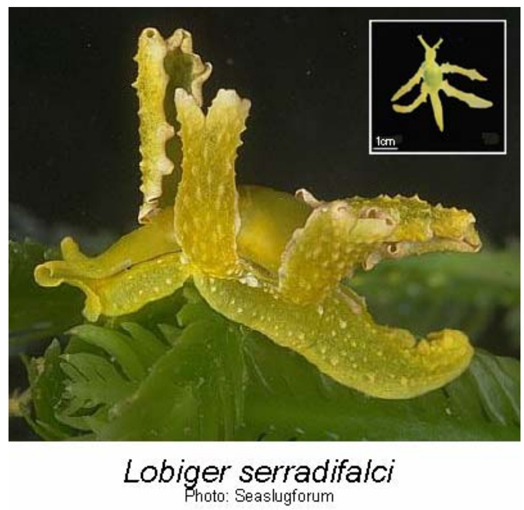


Fig.5.d: *Lobiger serradifalci*.

Elysia subornata, a Caribbean species lacking a shell; it feeds only on *Caulerpa* species by causing incisions with the radula - incised algal thalli rapidly become necrotic and die. The grazing rates correspond to the destruction of 5-6cm/day of frond at 21°C; this is 2-11 times higher than those recorded for the Mediterranean ascoglossan species ([5.3]Thibaut & Meinesz, 2000). Being a tropical species it no longer reproduces at 17°C and dies at 15°C. Dietary switching is possible on some Mediterranean caulerpales but feeding on other algae and sea grass is unlikely. According to Thibaut et al., ([5.6]2001) only 30% of the *E.subornata* individuals tested survived when fed a diet made up of *C.prolifera* exclusively, whereas 100% of individuals tested survived when fed with *C.racemosa*.

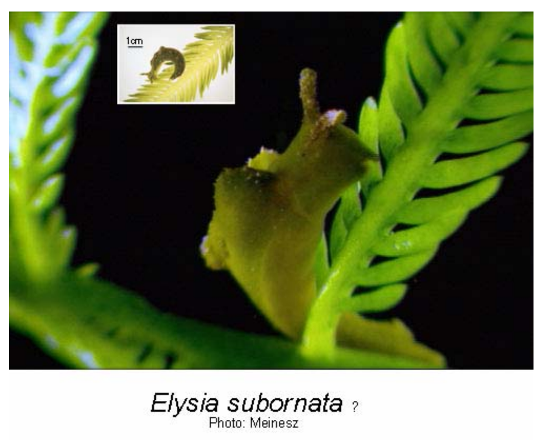


Fig.5.e: *Elysia subornata*.

Incisions were observed on the Ulvaceae, *Enteromorpha compressa* and *Ulva* sp. but the ascoglossans were unable to survive beyond 40 days when exclusively fed on these algae. Only this species is able to ingest and store resistant chloroplasts (kleptoplasty) to allow individuals to survive if food is lacking ([5.7]Clark et al., 1990); this explains the high degree of stenotrophy of *E.subornata* towards *Caulerpa* sp. Another important aspect speaking for *E.subornata* is its benthic larval development ([5.4]Clark et al. 1979) allowing it to quickly establish locally dense populations.

Unless species are found that are more temperature tolerant, molluscan species that would not survive the winter period in the Mediterranean Sea where temperatures are below 15°C, and where feeding and reproduction are only significant above 20°C, would require breeding to be performed under laboratory conditions prior to the release window stretching from May to October. With feeding rates of around 5cm of frond per day (*E.subornata*), still more than 1000 slugs/m^2 of *C.taxifolia* are required to obtain a significant effect on a dense colony with approximately 5000 fronds/m^2 ([5.8]Meniesz et al., 1995). Regardless of planktothrophic larval stage (*Oxynoe*, *Lobiger*) or lower thermal viability (*Elysia*), yearly treatments may be required to maintain a sufficient level of predation on the alga and thus reduce its biomass in the long run ([5.9]Coquillard et al., 2000).

Already now, biologists have bred thousands of snails, which include also the tropical variant in laboratories around *Nice* (France). Teams of scientists are waiting for permission from the French authorities to unleash the snail army. But critics fear that the remedy may introduce new bio-troubles. As recommended by the International Council for the Exploration of the Sea (ICES) and the Food Administration Organization's (FAO) guidelines on biological control or the introduction of species ([5.10]FAO, 1997; [5.11]ICES, 1997) this study on nonnative ascoglossans from the Mediterranean Sea should not be undertaken in open sea. The French Ministry of Environment shares the same attitude. As far as the risks associated with the introduction of non-native species, four of these are identified below:

- switching to non-target species - unlikely in the case of *E.subornata* as it is strictly stenotrophic;
- introduction of pathogens - with species imported from the Caribbean (*Elysia*) no disease transfer was observed even after one year of quarantine ([5.12]Pierce et al. 1999);
- competition with indigenous Mediterranean ascoglossan - theoretically possible, but due to thermal limitations in their reproductive cycle, annual intervention is necessary; thus limiting competition to the time window an eventual trial is running;
- spreading across the Mediterranean - anti-Lessepsian migration is possible (into the Red Sea and eastern Atlantic).

Meinesz, an advocate of the operation, is convinced that it is safe. Due to its stenotrophic spectrum, dietary switching is not very probable. Furthermore, releasing it in the spring will result only in a temporary invasion as it will die out during the cooler winter months.

5.b Vectors aiding in the spread of C.taxifolia: Although natural (wind & ocean currents) vectors aid in its distribution, the main dispersal agent spreading *C.taxifolia* across the globe is human-mediated. The aquarium trade in particular, is the most likely source of introduction to the Australia, Oceania, and the Americas. Similar species are sold on the Internet, although far too often the exact species traded is not even known.

As outlined before, dissemination essentially takes place by fragmentation. All sites of infestations far from the introduction area are either close to a harbor, common sites of leisure activity (anchored), or extensively harvested fishing areas (compare fig.5.h, [5.13a]Creese et al., 2004). A fact that confirms dispersion of plant fragments attached to boat anchors, fishing gear (including those of commercially operating enterprises) and other equipment. Although unlikely, *Caulerpa* can be introduced in ballast water or on vessel hulls (NZFMB)[5.14].

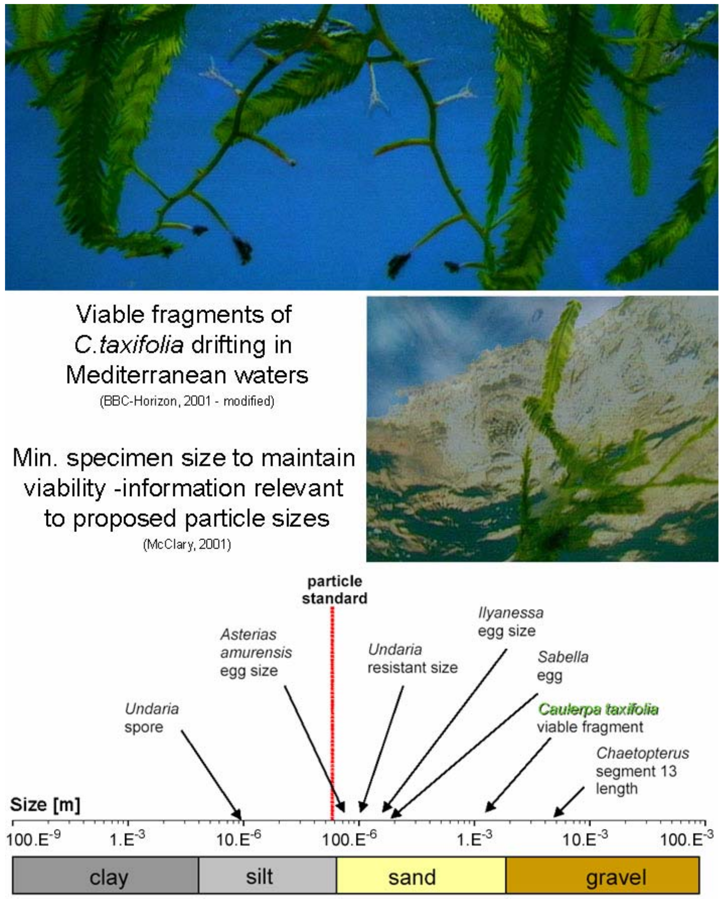


Fig.5.f: Viability size of floating *C.taxifolia* fragment.

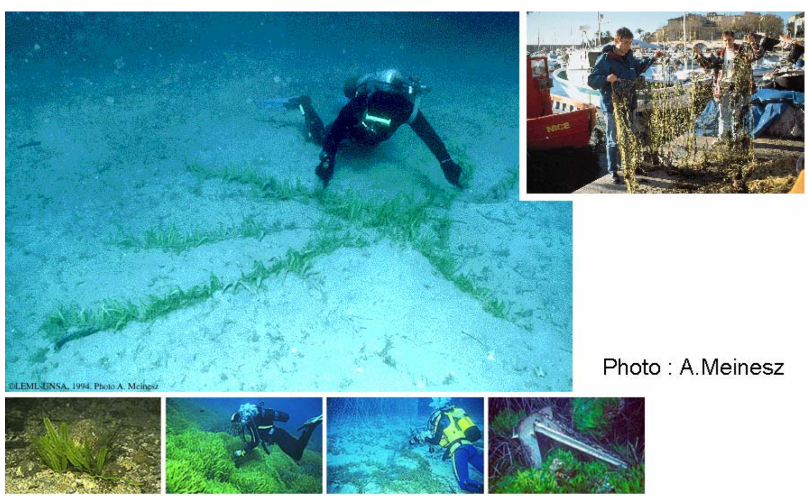
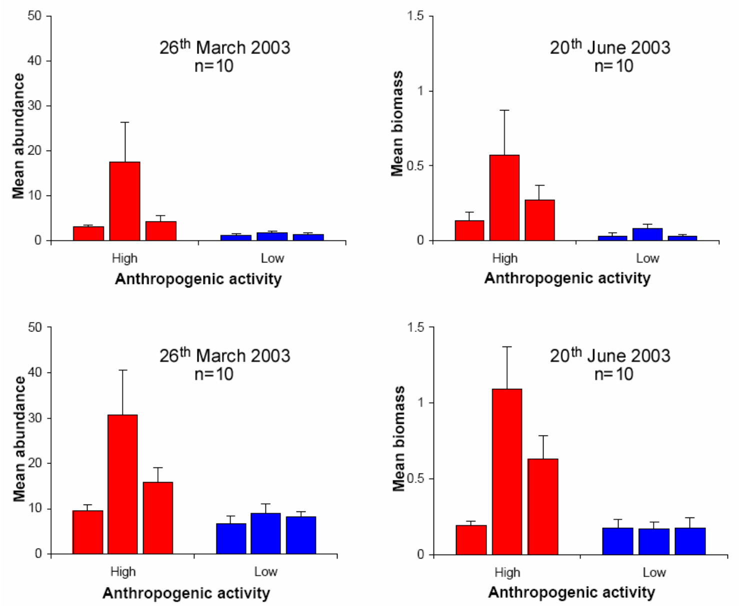


Fig.5.g: Vectors of dispersion.

However, even a barely visible piece can regenerate a new plant (fig. 5.f). At 25°C, formation of a whole small plant including fronds, stolons and rhizoids, from a cutting as small as a few mm in size ([5.34]McClary, 2001), occurred within 10 days following a similar pattern of regeneration ([5.13]Zuljevic & Antolic, 2002). This has rendered all but useless any attempt to eradicate the algae, either by hand or by using the underwater equivalents of plows. The spreading of the alga is facilitated by fishing activity, in particular by bottom trawlers and trammel nets. Fishermen are themselves strongly affected by the spreading, not only because of the decrease in valuable fish, but also because the massive presence of the alga interferes with the use of the gear. In the shallow, bottom trawling is forbidden by EU and Italian laws, but illegal fishing is not uncommon and so a trawler can easily transport algal fragments for several kilometers on a single day ([5.15]Relini et al., 2000). The same authors go even further by attesting that fishermen harm themselves with these fishing practices in two ways:

i) First, because of the qualitative change in catch composition, commercially important species such as *Pagellus erythrinus*, *Pagrus pagrus*, and *Solea lascaris* decrease and may ultimately disappear when the sea grass is replaced by the green alga.
i) And secondly, because the massive presence of *C.taxifolia* interferes with fishing activities. In particular, the large quantity of algal fronds affects the deployment and performance of the gear.

Fig.5.h: Anthropogenic influence.

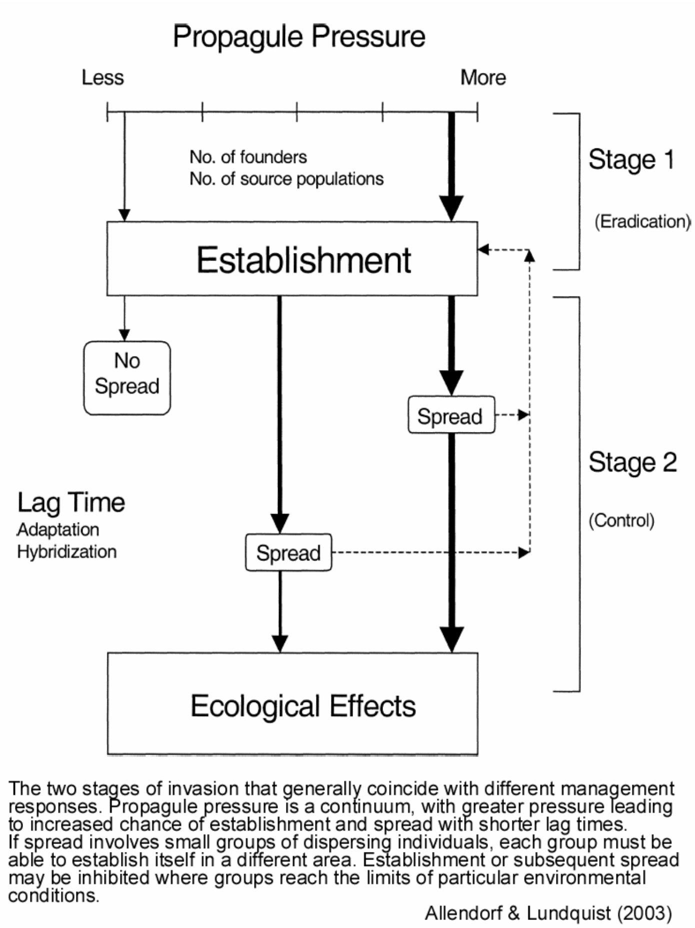


Mean (+s.e.) abundance (left) and biomass (g dry weight) (right) per quadrat, of *C.taxifolia* fragments sampled in *Lake Conjola* at three locations with high anthropogenic activity and three locations with low anthropogenic activity, on two sampling occasions.
(adapted from Creese et al., 2004)

5.c Countermeasures - What has been done so far? In Italy, except for a few eradication attempts made at the onset of the invasion, and Tunisia alike, no control strategy has been established ([5.16]Thibaut & Meinesz, 2002). Many methods to control this plant have been tested throughout the Mediterranean. Some have tried to tear up the patches of algae but one torn leaf that gets away can generate a whole new outbreak. Divers have used pumps to pull out the plant but it seems to regenerate in the same place at a rate quicker than its original growth rate. Other eradication methods include poison, smothering the algae with a cover that lets in no light, and using underwater welding devices to boil the plant.

Manual uprooting has been executed by trained and motivated divers. It is a solution for small algal patches measuring a few square meters, but even then it is not 100% effective. Sometimes there is re-growth and the operation has to be repeated. In France, regular control of the alga only occurs in the waters of the national park of *Port-Cros*, where control efforts (manual removal) have been performed annually since 1994. Fifteen tiny isolated colonies have been successfully eradicated ([5.17]Riera et al, 1994; [5.16]Thibaut & Meinesz, 2002). This technique is unfeasible, and tends to be a lost cause from the outset in-depth growth, guaranteed re-growth and exorbitant cost. It could have been effective in 1991 when only a few hectares were colonized.

Fig.5.i: Development of *C.taxifolia* species invasion ([5.35]Allendorf & Lundquist, 2003).



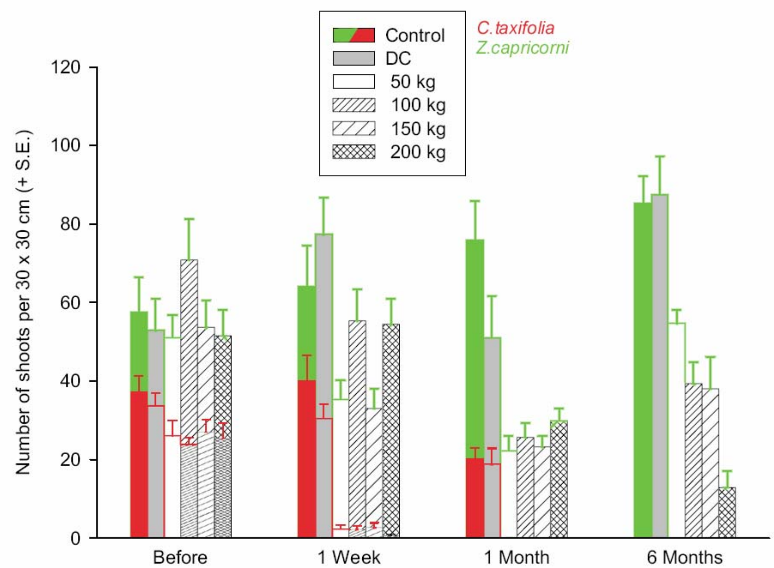
Physico-chemical elimination procedures were considered and tested either in an aquarium or at an experimental site. These involve certain chemicals, cross-ionic dialysis, vacuum hoses, airlift sediment suckers, suction pumps, dry ice, ultrasound, hot water jets, etc. Although not very efficient with larger patches, these methods can be applied in areas with smaller infestations; e.g. being smaller in extension, annual control measures in Croatia have been implemented by covering isolated colonies with black plastic sheets and removing the alga with a suction pump.

- Smothering with rock salt (sodium chloride, NaCl) has shown partial success in that it kills the *C.taxifolia* assimilators, but the health of the rhizoids imbedded in the soft sediment has yet to be determined (Millar & Talbot, 2002). Smothering techniques are currently being tested in Australian's West Lakes and may prove to be a suitable management tool for small, isolated patches of the weed. Fresh water is an option when *C.taxifolia* is encountered in lagoons that can be sealed off. Indeed Australian authorities investigated the possibility whether the affected area could be turned into a fresh water body. The weed dies in fresh water with salinity levels less than 10 parts per thousand. However, trials performed with NaCl in *Lake Macquarie* showed that a concentration of 50kg/m^2 of salt could remove all *C.taxifolia* fronds for a period of at least 6 months if applied during autumn (fig.5.j, [5.13a]Creese et al., 2004).
- Copper (Cu): is known to be toxic not only to plants, but also to the entire underwater ecosystem. Since the first discovery in 1992, Spanish authorities have been shown to slow down the spread of the alga by using Cu-electrodes, and Cu-salts (ions). Similar efforts were undertaken by French authorities in the *Port Cros* region since 1994, by applying cloths soaked in copper salts, ([5.16]Thibaut & Meinesz, 2002).
- Chlorine: As part of the eradication effort in *Agua Hedionda* and *Huntington Harbour* (California-USA) each patch of *Caulerpa* was covered with a heavy plastic tarpaulin, sealed to the bottom at the edges and fitted with a small port on top that allowed the introduction of liquid chlorine into the mud

and water. Chlorine not only killed *C.taxifolia* but also any other life form present under the tarps. The tarpaulins were left in place to prevent re-growth below ground material and other parts that may not have been fully treated, while preventing loss of herbicide to lagoon waters ([5.19]Withgott, 2002). Long-term monitoring will be necessary at least for the next five years on a monthly bases to assure complete eradication. Financially, the eradication program at the Californian site (*Agua Hedionda* Lagoon) in the first two years of the program amounted to approx. U\$1.9·E^6. With the amount of *Caulerpa* being reduced as work progressed, surveillance has replaced treatment. Therefore another U\$800·E^3/year has been estimated as a minimal requirement to declare the eradication program a success ([5.20]Merkel & Woodfield, 2003).

- Algacide have proven to be ineffective.

Fig.5.j: NaCl treatment of *C.taxifolia*.



Mean number of fronds of *C.taxifolia* and *Zostera capricorni* in plots that were either not salted (close control (Control) & distant control (DC)), or salted at concentrations of 50, 100, 150 or 200 kg/m^2. Data for each treatment are averaged across replicates, plots and sites. The experiment was initiated in March 2002 in Lake Macquarie. (adapted from Creese et al., 2004)

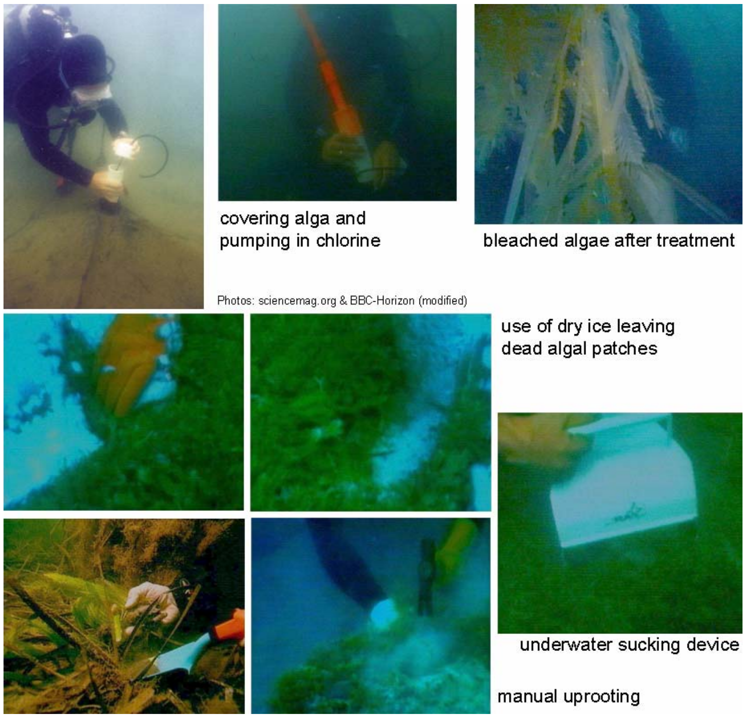


Fig.5.k: Manual Control of *C.taxifolia*.

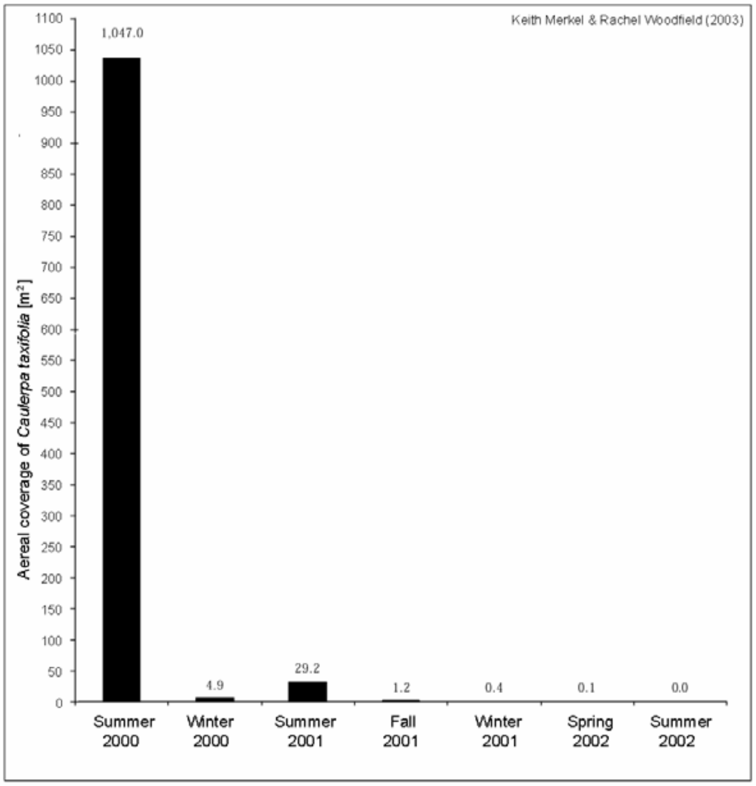


Fig.5.l: Eradication Success in California.

Since these methods do not meet one or more of the criteria (effectiveness, absence of re-growth after one month, non-dispersal of cutting, absence of secondary effects on other systems), the only feasible strategy is not one of total eradication but rather one of slowing down the rate of spread by eradicating small, isolated patches through a combination of various techniques.

Biological Methods may well be the only feasible solution in the future for the Mediterranean. The evaluation of laboratory trials involving Mediterranean specialist grazing exclusively on this species (e.g. Mollusca, Opisthobranchia) showed that indigenous species are inefficient to halt its spread. On the other hand, the tropical counterpart *Elysia subornata* provides some hope, if a cold-resistant strain of this species could be cultivated ([5.16]Thibaut & Meinesz, 2002). According to the optimal foraging theory ([5.21]Hugues, 1980), total eradication of all the *Caulerpa* spp. by biological control methods is ecologically not possible. Any practical applications of these methods, already being studied, must be accompanied by the necessary authorization and precautions so that the remedy itself, should it prove effective, does not cause further unforeseen upsets to the already stressed ecosystem. There are no universal and reliable rules, which can predict whether a biological control organism will be effective ([5.22]Carlton, 1997).

1. Genetic research, which is advancing in leaps and bounds, could also provide solutions for local elimination and prevention. Though, one should keep in mind, that once released, a genetically modified organism can never be withdrawn any more!
2. Mapping *C.taxifolia*'s expansion in the Mediterranean Sea is necessary for a number of reasons (even though, as the extent of colonization increases, mapping precision decreases, [5.23]Vaugelas et al., 1999):
   (i) to follow its progression from one year to the next;
   (i) to assess its environmental impact and analyze the regression of out-competed species;
   (i) to estimate *C.taxifolia* biomass and other related biological data;
   (i) to calibrate computer models to simulate its spread;
   (i) to describe the situation to decision-makers for possible control measures.
   Eventually, the left over smaller patches could be treated with physico-chemical means.

The present state of the Mediterranean invasion is critical and any attempt of eradication of the alga seems useless. A realistic option would be to preserve the biodiversity of selected sanctuaries against the invasion by regular control of new *C.taxifolia* recruitment. In addition, modeling the spread can help decision makers with their choice of strategy. A selection of simulation models taking into account the biology of *C.taxifolia*, the seasons, and the spatial characteristics of the Mediterranean are used. Modeling results are accurate over 4-5 year time periods ([5.24]Aussem & Hill, 1999; [5.25]Lemée et al., 1997).

5.d. Preventive Methods: Emptying aquarium contents facilitates short distance spread as fragments transported by currents quickly establish newly colonized sites; these are usually harbors, marinas and other places where boats anchor ([5.26]Ribera & Boudouresque et al 1995). The long distance spread of *C.taxifolia* can be the result of cleaning anchors and fishing nets as well as ballast water ([5.27]Meinesz, 1992; [5.28]Sant et al., 1996). Hence, being asexually in reproduction, this is important information for resource management and conservation. The uppermost aim though, must be prevention, and this not only involves local authorities but every individual that has to do directly or indirectly with this algae. Thus the guidelines listed herein, should be strictly followed:

1. Home-Aquaria: As alternatives are available, every owner of a salt-water aquarium should refrain from using this seaweed! Salt-water aquaria and other contents should never be emptied into or near any gutter, storm drain, creek, lagoon, bay, harbor, or the ocean. Aquarium water should be disposed off only in a sink or toilet connected to a wastewater treatment plant. Rock and other solid material from an aquarium should be disposed off in a trash-can. *C.taxifolia* from an aquarium (and anything it is attached to), should be either dried out for several days under the sun or placed in a plastic bag, put in a freezer for at least 24 hours, and then disposed in a landfill.
2. Fishing: If any seaweed suspected to be *C.taxifolia* is found on fishing gear it should be removed, carefully bagged (definitely not thrown back into the sea, since even a small fragment has the potential to regenerate into a new plant), and reported. In order to prevent new infestations and comply with the law, *C.taxifolia* should not be purchased, sold, or distributed. The legal provisions adopted in France and Catalonia (ban on buying, selling, transporting and storing of *C.taxifolia*) should be adopted by all countries around the Mediterranean.
3. Boat: Long-distance spread should be avoided by informing owners of private vessels of the need to check and clean their anchors, trailers, rudders, after mooring in contaminated areas. Like with the

above, algal fragments should be removed, carefully bagged (not thrown back into the sea or carried to a new location), dumped into a trash-can and reported to the authorities. Mooring should be prohibited in highly contaminated areas, all national and international regulations and legislation on the introduction of species will have to be adapted.

4. Water Sports: Sun-lovers, snorkellers, divers, and fishermen should be instructed to inform their local authorities and environmental services each time they sight new patches or populations of *C.taxifolia*. Such information is essential for the continued monitoring and its spread as well as the adoption of any necessary measures and must also include active involvement of the tourist industry.
5. Everybody's help is needed! Your help is critical to the success of containing and prevent further infestation by this destructive marine seaweed (refer to contact details in Chapter 6).

5.e. *Caulerpa* and the Law: Boudouresque & Verlaque ([5.29]2002) recommend that steps be taken at once to slow down the rate of introduction of non-native species. In particular, it seems necessary to implement national legislation, to set up quarantine conditions (aquaculture), to control the flow of ballast waters and the aquarium trade and to ban all species, which prove to be invasive in other parts of the world ("black list" or "dirty list"). Subsequently, it will be necessary to move from a "dirty list" to a "clean list" approach: only the species mentioned on the "clean list" will be allowed to be imported. In addition, it is clear that laws should be enforced and that particularly lax practices should be stopped.
At the same time, the spread of an aquarium strain of *C.taxifolia* in the Mediterranean has led several governments (Australia, France, Spain and USA) to ban its use in the aquarium trade in order to prevent it from escaping to new geographical areas (Jousson et al., 1998).

EU: In a decree dated 4th of March 1993, the French Minister for the Environment and the State Undersecretary for the Sea banned the offering, the sale, buying, use and dumping into the sea of all or parts of the specimens of the algae *Caulerpa taxifolia*. Collection and transport of the algae are also subject to a system of authorization granted on presentation of a well-grounded request. The appearance and spread of *C.taxifolia* are covered at Mediterranean level by the two provisions of article 13 of the Barcelona convention's Protocol on Specially Protected Areas which was adopted in 1996 but has not yet come into force (entitled: The introduction of non-indigenous or genetically modified species).
AUS: The risk of an introduction of non-native *C.taxifolia* to Australian waters has been recognized by the Australian Quarantine and Inspection Service with the implementation of an import ban of the species in 1996 ([5.30]Schaffelke et al., 2002). The alga was listed as a Noxious Species by the parliament of New South Wales (NSW) on 1st of October 2000; it cannot be bought, sold, traded, or kept in an aquarium in NSW ([5.18]Millar & Talbot, 2002).
NZ: The New Zealand government put the aquaria strain of *C.taxifolia* to the list of species on the Plant Pest Accord for surveillance of retail outlets by Regional Councils. The so-called aquarium strain of *C.taxifolia*, referring to the Mediterranean invasion, is on the interim species list to trigger a marine pest incursion emergency response in Australia ([5.31]Schaffelke et al. 2002).
USA: Assembly Bill 1334 (Harman), signed into law by the Californian Governor in September 2001, prohibits the possession, sale, and transport of *C.taxifolia* throughout that state. This bill also establishes the same restrictions on several other species of the genus *Caulerpa* that are similar in appearance to *C.taxifolia* and that are believed to have the ability to become invasive. Furthermore, the importation, interstate sale (including Internet sale), and transport of the Mediterranean strain (i.e., aquarium strain) of *C.taxifolia* is prohibited under the federal Noxious Weed Act ([5.32]1999) and the federal Plant Protection Act ([5.33]2003).

6. Conclusion, Contact Details, Web-links & Bibliography

6.a Conclusion: As it establishes itself as an extremely successful in a wide range of environmental conditions, the aquarium strain of *Caulerpa taxifolia* is a highly invasive green alga that has shown to cause major ecological and economic damage in the north-western Mediterranean. If the spread continues, moving as far as eastward into the Black Sea or the Red Sea, consequences could be even worse than those registered so far. Likewise, no one can rule out the possibility of the algae traveling through the Straits of Gibraltar and spreading up or down the Atlantic coastline of Africa or Europe. As a result the Global Invasive Species Specialist Group has grouped *C.taxifolia* among the 100 most "Worst Invasive Alien Species" ([6.0]ISSG, 2004).

The fact that researchers have found *C.taxifolia* off the coast of California (USA) and along the south coast of South Australia, vectors such as ballast water of container ships (although less likely) or the drainage of home salt-water aquaria must have contributed to the spread of this algae into these parts of the world.

Therefore, any invasion of new territory must be tackled immediately and action plans applicated to prevent any further spread. This seaweed has a devastating ecological and economic impact in the Mediterranean. It has formed dense carpets, out-competed native seaweeds and sea-grasses and displaced invertebrates. Such carpets can also cause sediment anoxia, which affects the infauna. Chemical analyses and feeding trials have demonstrated that the alga contains toxins that deter herbivores, including fish. Where *C.taxifolia* has invaded, species diversity and abundance is reduced, resulting in substantial losses in fisheries production. In addition, the establishment of *C.taxifolia* is reported to have harmed tourism and pleasure boating, and recreational diving, and commercial fisheries.

Humans bear the prime responsibility for most of the allochthonous species introduction; some of which are harmful or represent a cause of environmental concern. The scale of the areas of Mediterranean colonization by *C.taxifolia*, the threat, which they pose to eco-diversity, resources and certain activities (scuba diving, tourism in general, and fishing industry) demand immediate action. For the time being, the choice of available means of removal is unfortunately very limited. This introduced species has shown to radically alter the structure and function of native ecosystems, causing a decrease in biodiversity ([6.1]Ceccherelli & Cinelli, 1997; [6.2]Piazzi & Cinelli, 2000). Although theory predicts that communities rich in species should be less susceptible to invasion ([6.3]Rejmánek, 1989; [6.4]Case, 1990; [6.5]Law & Morton 1996; [6.6]Stachowitcz et al., 1999; [6.6]Prieur-Richard & Lavorel, 2000), *C.taxifolia* has proven to be an opportunist whenever an ecosystem is out of balance. Indeed, it is a kind of "ecological roulette", as coined by Carlton and Geller ([6.8]1993) what we as members of *Homo sapiens* are doing with our environment.

Though the alga is not a "killer", the damage caused to our environment is already manifest, including the ecological consequences for the fauna and flora - and finally for humans who exploit them - can be very grave. The years of inaction produced and will continue to produce, in any case, enormous costs for society.

Therefore our appeal to You: Do not spread *Caulerpa* any further for any purpose.
Report any suspected sightings of *Caulerpa* to the nearest authorities!

6.b Contact Details - for further information or reporting *C.taxifolia* sightings in your area or if you find this seaweed do not help it to spread, but contact:

EU: Laboratoire Environment Matrin Litoral, CNRS UMR "DIMAR" 6540
University of Nice-Sophia Antipolis, Faculty of Sciences - c/o: Prof. Alexandre Meinesz
F-06108 Nice – **France**
phone.: +33.(0)492.076.846
fax.: +33.(0)492.076.849
e-mail: meinesz@unice.fr or gravez@com.univ-mrs.fr
http://www.unice.fr/LEML

Institute "Ruder Boskovic" Center for Marine Research - c/o: Dr. N. Zavodnik or A. Jaklin

Giordano Paglia 5
HR-52210 Rovinj – **Croatia**
phone: +385(052).811.544 or 804 700
fax.: +385(052).813.496
e-mail: cmrr@more.cim.irb.hr
http://more.cim.irb.hr

Institute of Oceanography and Fisheries - c/o: Dr. Boris Antolic, Dr. A. Span or A. Zuljevic
I Mestrovica 63 - P.P. 500
HR-21000 SPLIT – **Croatia**
phone: +385(021).358.688
fax.: +385(052).358.650
e-mail: office@izor.hr
http://www.izor.hr

Università di Pisa, Dipartimento di Scienze dell'Uomo e dell'Ambiente - c/o: Prof. Francesco Cinelli
Via A. Volta 6
I-56127 Pisa – **Italy**
phone: +39(050).221.9011
fax: +39(050).050.49694
e-mail: fcinelli@scamb.unipi.it
http://www.unipi.it

Istituto di Anatomia Comparata dell'Università di Genova - c/o: Dr. Giorgio Matricardi
ViaIe Benedetto XV n.5
I-16132 Genova – **Italy**
phone: +39(010).353.8053
fax: +39(010).353.8047
e-mail: :matric@unige.it
http://www.unige.it

Università degli studi di Palermo, Dipartimento di Scienze Botaniche - c/o: Dr. Carla Fradà Orestano
Via Archirafi 38
I- 90123 Palermo - **Italy**
phone: +30(091).623.8001
fax: +39(091).623.8203
e-mail: versace@unipa.it
http://www.unipa.it

Centro de Estudios Avanzados de Blanes - c/o: Dr. Enrique Ballesteros Segarra
Camí de Santa Bàrbara s/n
E-17300 Blanes – **Spain**
phone: +34(972).33.61.01
fax: +34(0)972.33.78.06
e-mail: kike@ceab.csic.es
http://www.ceab.csic.es

Institute of Marine Science and Technology - c/o: Prof. Dr. Sukran Cirik
Haydar Aliyev Blv. No:10 Inciralti
TR-35340 Izmir – **Turkey**
phone : +90.(0232).278.5272 or +90.(0232).278.6515 or +90.(0232).278.6525
fax: +90.(0232).278.5082
e-mail: cirik@Imst.deu.edu.tr
http://www.deu.edu.tr

FEZZ - c/o Artur Strauss
Korbinianstrasse 45
D-80807 Munich – **Germany**
e-mail: ars.ase.ts@t-online.de
http://home.t-online.de

AUS & NZ: New South Wales Fisheries - c/o: Dr Alan Millar
Royal Botanic Gardens Sydney
Mrs Macquarie Rd
SYDNEY NSW 2000 – **Australia**
phone: +61(02).4916.3877or +61(02).9527.8411 or +61(02).4916.3811
e-mail: pests@fisheries.nsw.gov.auor alan_millar@hotmail.com or tsadmin@fisheries.nsw.gov.au
http://www.fisheries.nsw.gov.au/index.html

Chief Technical Officer - Marine Biosecurity / Ministry of Fisheries
PO Box 1020
Wellington - **New Zealand**
phone: 0800 INVADERS (468 233)
e-mail: biosecurity@fish.govt.nz
http://www.fish.govt.nz

USA: Merkel & Associates Inc. - c/o: Dr. Rachel Woodfield
3944 Murphy Canyon Road, Suite C 106
San Diego, CA 92123 – **USA**
phone: 858-560-5465
e-mail: rwoodfield@merkelinc.com
http://swr.ucsd.edu/hcd/CAULERPA.htm

National Marine Fisheries Service Southwest Regional Office - c/o: Dr. Bob Hoffman
01 West Ocean Boulevard, Suite 4200
Long Beach, CA 90802 – **USA**
phone: +1(562).980.4043
fax: +1(562).980.4092
e-mail: or wpaznokas@dfg.ca.gov
http://bonita.mbnms.nos.noaa.gov/welcome.html
http://www.caulerpa.cjb.net/ or http://www.sccat.net

Calif. Dept. of Fish & Game South Coast Region - c/o: William Paznokas
4949 Viewridge Drive
San Diego, CA 92124 – **USA**
phone: +1(858).467.4218
fax: +1(858).467.4299
e-mail: wpaznokas@dfg.ca.gov
http://swr.nmfs.noaa.gov

6.c Literature The information contained herein is based on the following literature:

PART-I

[1.0]ISSG, 2004; Invasive Species Specialist Group of the World Conservation Union (IUCN); (http://www.issg.org/database/species/search.asp?st=100ss&fr=1&sts=#);

[1.1] Jousson O., et al. 2000; *Invasive Alga Reaches California*; Nature, Vol. 408: 157;

[1.2] Meinesz A., 1999; *Killer Algae: The True Tale of a Biological Invasion*; Univ. of Chicago Press (IL) – USA;

[1.2a] NIMPIS, 2002; *Caulerpa taxifolia species summary*; National Introduced Marine Pest Information System (Eds: Hewitt C.L., Martin R.B., Sliwa C., McEnnulty, F.R., Murphy, N.E., Jones T. & Cooper, S.); (http://crimp.marine.csiro.au/nimpis);

[1.2b] Creese R.G., Davis A.R. Glasby T.M.; 2004; *Eradicating and preventing the spread of the invasive alga Caulerpa taxifolia in NSW*; NSW Fisheries Final Report Series No.: 64; NSW Fisheries, Cronulla – AUS;
[1.3] Chisholm J.R.M., Fernex F.E., Mathieu D., Jaubert J.M., 1997; *Wastewater, Seagrass Decline and Algal Proliferation on the Côte d'Azur*; Marine Pollution Bulletin, Vol.34, Issue2:78-84;
[1.4] Meinesz A., 1992; *Modes de dissémination de l'algue Caulerpa taxifolia introduite en Méditerranée*; Rapp. Comm. internation. Explor. sci. Médit., 33:B-44;
[1.6] Boudouresque C.F., Meinesz A, Ribera M.A., Ballesteros E., 1995; *Spread of the green alga Caulerpa taxifolia (Caulerpales, Chlorophyta) in the Mediterranean: possible consequences of a major ecological event*; Sci.Mar.59 (Suppl.1):21-29;
[1.7] Schaffelke B., Murphy N.; Uthicke S., 2002; *Using genetic techniques to the sources of the invasive alga Caulerpa taxifolia in three new locations in Australia;* Marine Pollution Bulletin, Vol.44:204-210;
[1.8] Paul V.J. 2002; *Chemical Ecology of Caulerpa spp. with an Emphasis on Invasive Caulerpa taxifolia*: University o.Guam Marine Lab. - USA; (http://sgnis.org/update/ctax.htm);
[1.9] Pesando D., Lemée R., Ferrua C., Amade P., Girard J.P., 1996: *Effects of caulerpenyne, the major toxin from Caulerpa taxifolia on mechanisms related to sea urchin egg cleavage*; Aquatic Toxicology, Vol.35:139-155;

PART-II

[2.0] Integrated Taxonomic Information System (ITIS),data retrieved online (4th Nov. 2004); (http://www.itis.usda.gov/index.html);
[2.1] Raven P.H., Evert R.F. Eichhorn S.E., 1992; *Biology of Plants* 5th ed.; Worth Publishers, NY - USA;
[2.2] Dumay O., Pergent G., Pergent-Martini C., Amade P., 2002 *Variations in Caulerpenyne contents in Caulerpa taxifolia and C.racemosa*;J.o. Chemical Ecology Vol.28;
[2.3] Mandoli D., Olmstead R., Mishler B., Boore J.L., Smith A.R., Renzaglia K., Wolf P., Donoghue M.J., O'Kelley C., 2002; *Toward Resolution of the Fuzzy Nodes in Green Plant Phylogeny;* see deep green plant phylogenetics.pdf: http://ucjeps.berkeley.edu/TreeofLife/poster.php;
[2.4] Strassburger E., 1998; *Lehrbuch der Botanik* 33rd ed.; Fischer Verlag; Stuttgart – FRG;
[2.5] Silva, P.C., 2002; *Overview of the Genus Caulerpa*; Herbarium of the Univ. of California, Berkeley (CA) – USA; (http://sgnis.org/publicat/silv2002.htm);
[2.6] Meinesz A., Benichou L., Blachier J., Komatsu T., Lemée R., Molenaar H., Mari X., 1995; *Variations in the structure, morphology and biomass of Caulerpa taxifolia in the Mediterranean Sea*; Bot. Mar. 38, 499-508;
[2.6a] Creese R.G., Davis A.R. Glasby T.M.; 2004; *Eradicating and preventing the spread of the invasive alga Caulerpa taxifolia in NSW*; NSW Fisheries Final Report Series No.: 64; NSW Fisheries, Cronulla – AUS;
[2.7] Boudouresque C. F., Meinesz A., Ribera M. A., Ballesteros E., 1995; *Spread of the green alga Caulerpa taxifolia (Caulerpales, Chlorophyta) in the Mediterranean: possible consequences of a major ecological event*; Sci.Mar.59 (Suppl.1):21-29;
[2.8] Benzie J.A.H., Ballment E., Chisholm J.R.M., Jaubert J.M., 2000; *Genetic variation in the green alga Caulerpa taxifolia*; Aquat.Bot. 66:131-139;
[2.11] Meinesz A, Hesse B., 1991; *Introduction et invasion de l'algue tropicale Caulerpa taxifolia en Méditerranée nord-occidentale*;Oceanol Acta 14:415-426
[2.12] Chisholm JRM, Dauga C, Ageron E, Grimont P. 1996; *Roots' in mixotrophic algae*; Nature 381:382
[2.13] Jaasund, E., 1977; *Marine algae in Tanzania* VIII. Bot. Mar. 20:509-520;
[2.14] Coppejans E., Beekman T., 1990; *Caulerpa (Chlorophyta, Caulerpales) from the Kenyan coast*; Nova Hedwigia 50:111-125;
[2.15] Coppejans E., Prud'homme van Reine W.F., 1992; *Seaweeds of the Snellius II Expedition (E. Indonesia): the genus Caulerpa (Chlorophyta Caulerpales)*; Bull. Seanc. Acad.roy.Sci.Outre-mer Meded.Zitt.K.Acad. Overzeese Wet. 37:667-712;
[2.16] Calvert H.E., Dawes C.J., Borowitzka M.A., 1976; *Phylogenetic relationships of Caulerpa (Chlorophyta) based on comparative chloroplast ultrastructure*; J.o. Phycol. 12:149-162;
[2.17] Silva P.C., Menez E.G., Moe R.L., 1987; *Catalog of the benthic marine algae of the Philippines*; Smithsonian Contrib.Mar.Sci. 27:1-179;
[2.18] Chisholm J.R.M., Jaubert J.M., Giaccone G., 1995; *Caulerpa taxifolia in the northwest Mediterranean: introduced species or migrantfrom the Red Sea?*; C.R.Acad.Sci. Paris, Sciences de la vie 318:1219-1226;
[2.19] Olson J.L., Valero M., Meusnier I., Boele-Bos S., Stam W.T., 1998; *Mediterranean Caulerpa taxifolia and C.mexicana (Chlorophyta) are not conspecific*; J.o.Phycol. 34:850-856;

[2.20] Piazzi L., Balestri E., Magri M., Cinelli F. 1997; *The spread of the tropical alga Caulerpa racemosa (Forsskal) J.Agardh (Bryopsidophyceae, Chlorophyta) along the Tuscan coast (Italy)*; Cryptogamie Algol. 18:343-350;
[2.21] Verlaque M., Fritayre P., 1994; *Modifications des communautés algales méditerranéennes en présence de l'algue envahissante Caulerpa taxifolia (Vahl) C.Agardh*; Oceanol. Acta 17:659-672;
[2.22] deVilléle X., Verlaque M., 1995; *Changes and degradation in a Posidonia oceanica bed invaded by the introduced tropical alga Caulerpa taxifolia in the north western Mediterranean*; Bot.Mar.38:79-87;
[2.23] Bellan-Santini D., Arnaud P.M., Bellan G., Verlaque M., 1996; *The influence of the introduced tropical alga Caulerpa taxifolia, on the biodiversity of the Mediterranean marine biota*; J.o.Mar.Biol.Assoc. U.K. 76:235-237;
[2.24] Ceccherelli G., Cinelli F., 1997; *Short-term effects of nutrient enrichment of the sediment and interactions between the seagrass Cymodocea nodosa and the introduced green alga Caulerpa taxifolia in a Mediterranean bay*; J.o.Exp.Mar.Biol. Ecol. 217:165-177;
[2.25] Piazzi L., Cinelli F., 2001; *Distribution and dominance of two introduced turf-forming macroalgae on the coast of Tuscany, Italy, n orth-western Mediterranean Sea in relation to different habitats and sedimentation*; Bot.Mar. 44(5):509-520;
[2.26] Piazzi L., Balestri E., Cinelli F., 1994; *Presence of Caulerpa racemosa in the north-western Mediterranean* Cryptogam., Algol. 15:183-189;
[2.27] Piazzi L., Balestri E., Magri M., Cinelli F., 1997; *Expansion de l'algue tropicale Caulerpa racemosa (Forsskal) J.Agardh (Bryopsidophyceae, Chlorophyta) le long de la côte toscane (Italie)*; Cryptogam., Algol. 18:343-350;
[2.28] Gambi M.C., Terlizzi A., 1998; *Record of large population of Caulerpa racemosa (Forsskal) J.Agardh (Chlorophyceae) in the Gulf of Salerno (Southern Tyrrhenian Sea, Italy)*; Biol.Mar. Mediterr. 5(1):553-556
[2.29] Modena M., Matricardi G., Vacchi M., Guidetti P., 2000; *Spreading of Caulerpa racemosa (Forsskal) J.Agardh (Bryopsidaceae, Chlorophyta) along the coasts of the Ligurian Sea*; Cryptogam., Algol. 21:301-304;
[2.30] Verlaque M., Boudouresque C.F., Meinesz A., Gravez V., 2000; *The Caulerpa racemosa Complex (Caulerpales, Ulvophyceae) in the Mediterranean Sea*; Bot. Mar. 43:49-68;
[2.31] Ceccherelli G., Piazzi L., Balata D., 2002; *Spread of introduced Caulerpa species in macroalgal habitats*; Journal of Experimental Marine Biology & Ecology, Vol.280:1-11;
[2.32] Jousson O, Pawlowski J, Zaninetti L, Meinesz A, Boudouresque C.F., 1998; *Molecular evidence for the aquarium origin of the green alga Caulerpa taxifolia introduced to the Mediterranean Sea*; Marine Ecology Progress Series, 172:275-280;
[2.33] Jousson O., Pawlowski J., Zaninetti L., Zechman F.W., Dini F., DiGuiseppe G., Woodfield R., Millar A., Meinesz A., 2000; *Invasive alga reaches California*; Nature 408:157;
[2.34] Benzie J.A.H., Ballment E., Chisholm J.R.M., Jaubert J.M., 2000; *Genetic variation in the green alga Caulerpa taxifolia*; Aquatic Botany, Vol.66:131-139;
[2.35] Olsen J.L., Valero M., Meusnier I., Boele-Bos S., Stam W.T., 1998; *Mediterranean Caulerpa taxifolia and C.mexicana (Chlorophyta) are not conspecific*; J.o.Phycol 34:850-856;
[2.36] Wiedenmann J., Baumstark A., Pillen T.L., Meinesz A., Vogel W., 2001; *DNA fingerprints of Caulerpa taxifolia provide evidence for the introduction of an aquarium strain into the Mediterranean Sea and its close relationship to an Australian population*; Marine Biology, 138:229-234;
[2.37] Marcabruno-Gerola F., 1968; *Regno Verde - Vol.2.* Fra.Fabri Editori, Milano – ITA;

PART-III

[3.1] Carlton J.T., 1985; *Transoceanic and interoceanic dispersal of coastal marine organisms; the biology of ballast water*; Oceanography and Marine Biology Annual Review 23:313-373;
[3.2] Ribera M.A., Boudouresque C.F., 1995; *Introduced marine plants, with special reference to macroalgae: mechanisms and impact*; In: Round F.E., Chapman D.J. (Eds.); Progress in Phycological Research, Vol. 11; Biopress Ltd Publ., pp.187-268;
[3.3] Williamson M., Fitter A., 1996; *The varying success of invaders*; Ecology 77 (6):1661-1666;
[3.4] Boudouresque C.F., Verlaque M., 2002; *Biological pollution in the Mediterranean Sea: invasive versus introduced macrophytes*; Marine Pollution Bulletin, Vol.44:32-38;
[3.5] Ashton P.J., Mitchell D.S., 1989; *Aquatic plants: patterns and modes of invasion, attributes of invading*

species and assessment of control programmes; In: Drake J.A., Mooney H.A., Di Castri F., Groves R.H., Kruger F.J., Rejmanek M., Williamson M. (Eds.), Biological Invasions: A Global Perspective. Wiley, Chichester, pp. 111-154;

[3.6] Pieterse A.H., 1990; *Introduction*; In: Pieterse A.H., Murphy K.J. (Eds.), Aquatic weeds; The Ecology and Management of Nuisance Aquatic Vegetation. Oxford University Press, Oxford, pp. 3-16;

[3.6a] Coquillard P., Gueugnot J., Hill D., Mazel C., Campos A.; 2001; *Caulerpa taxifolia Simulation over Cap Martin*; **Ec**osystem **Sim**ulation; Institut Supérieur d'Informatique de Modélisation et de leurs Applications – FRA; (http://www.isima.fr/ecosim/Welcome.html);

[3.7] Vaugelas J., Meinesz A., Antolic B., Ballesteros E., Belsher T., Cassar N., Ceccherelli G., Cinelli F., Cottalorda J.M., Orestano C.F., 1999; *Standardization proposal for the mapping of Caulerpa taxifolia expansion in the Mediterranean Sea*;Oceanologica Acta, Vol.22:85-94;

[3.8] Meinesz A., Cottalorda J.M., Chiaverini D., Garcia D., Thibaut T., DeVaugelas J., 2001; *Suivi de l'invasion de l'algue tropicale Caulerpa taxifolia en Méditerranée: Situation en France au 31 décembre 2000*; Laboratoire Environnement Marin Littoral, Université de Nice-Sophia Antipolis [ed.]. GIS Posidonie Publ.;

[3.9] Chisholm J.R.M., Fernex F.E., Mathieu D., Jaubert J.M., 1997; *Wastewater Discharge, Seagrass Decline and Algal Proliferation on the Côte d'Azur*; Marine Pollution Bulletin, Vol.34:78-84;

[3.10] Olivier G., 1929; *Etude de la flore marine de la Côte d'Azur*; Ann. Inst. oceanogr. Paris 8(3);

[3.11] Meinesz A., Laurent R., 1978;. *Cartographie et état de la limite inférieure de l'herbier de Posidonia oceanica dans les Alpes Maritimes (FRA)*; Bot. Mar. 21:513-526;

[3.12] Falconetti C., Meinesz A., 1989; *Charting the seaward limit of Posidonia meadows and of circalittoral biocoenoses along the coast of Monaco*; Oceanol. Acta 12:443-447;

[3.13] Chisholm J.R.M., Moulin P., 2003; *Stimulation of nitrogen fixation in refractory organic sediments by Caulerpa taxifolia (Chlorophyta)*; Limnol. Oceanogr., 48(2):787-794;

[3.14] Meinesz A, deVaugelas J., Cottalorda J.M., Caye C., Charier S., Commeau T., Delehaye L., Febvre M., Jaffrennou F., Lemée R., Molenaar H., Pietkiewicz D., 1994; *Suivi de l'invasion de l'algue tropicaleCaulerpa taxifolia devant les côtes francaises de la Mediterranée*; Lab. Environ. Marin Littoral, Univ. Nice-Sophia Antipolis & GIS Posidonie publication, Marseille, pp.121;

[3.15] Belsher T., Meinesz A., 1995; *Deep-water dispersal of the tropical alga Caulerpa taxifolia introduced into the Mediterranean* ; Aquatic Botany, Vol.51:163-169;

[3.16] Sant N., Delgado O., Rodriguez-Prieto C., Ballesteros E., 1994; *The effects of dessication on photosynthesis of Caulerpa taxifolia (Vahl) C. Agardh*; In: Boudouresque C.F., Meinesz A., Gravez V. (Eds.); "First International Workshop on Caulerpa taxifolia"; GIS Posidonie, Marseille, France, pp. 315-319;

[3.17] Delgado O., Rodriguez P.C., Gacia E., Ballesteros E., 1996; *Lack of severe nutrient limitation in Caulerpa taxifolia (Vahl) C Agardh, an introduced seaweed spreading over the oligotrophic northwestern Meditteranean*; Botanica Marina 39(1):61-67;

[3.18] Meinesz A, Hesse B., 1991; *Introduction et invasion de l'algue tropicale Caulerpa taxifolia en Méditerranée nord-occidentale*;Oceanol Acta 14:415-426

[3.19] Gnassia-Barelli M., Lemée R., Pesando D., Romeo M., 1995; *Heavy Metal Distribution in Caulerpa taxifolia from the North-Western Mediterranean*; Marine Pollution Bulletin, Vol.30;

[3.20] Meinesz A., Benichou L., Blachier J., Komatsu T., Lemée R., Molenaar H., Mari X., 1995; *Variations in the structure, morphology and biomass of Caulerpa taxifolia in the Mediterranean Sea*; Bot. Mar. 38:499-508;

[3.20a] Creese R.G., Davis A.R. Glasby T.M.; 2004; *Eradicating and preventing the spread of the invasive alga Caulerpa taxifolia in NSW*; NSW Fisheries Final Report Series No.: 64; NSW Fisheries, Cronulla – AUS;

[3.21] Zuljevic A., Antolic B., 2000; *Synchronous release of male gametes of Caulerpa taxifolia (Caulerpale, Chlorophyta) in the Mediterranean*; Sea. Phycologia, 39:157-159;

[3.22] Clifton K.E., Clifton L.M., 1999; *The phenology of sexual reproduction by green algae (Bryopsidales) on Caribbean coral reefs*; J.o.Phycol. 35:24-34;

[3.23] Silva, P.C., 2002; *Overview of the Genus Caulerpa*; Herbarium of the Univ. of California, Berkeley (CA) – USA; (http://sgnis.org/publicat/silv2002.htm)

[3.24] Raven P.H., Evert R.F. Eichhorn S.E., 1992; *Biology of Plants* 5th ed.; Worth Publishers (NY) - USA;

[3.25] Meinesz A, deVaugelas J., Benichou L., Caye C., Cottalorda J.M., Delehaye L., Febvre M., Garcin S., Komatsu T., Lemée R., Mari X., Molenaar H., Perney L., Venturini A., 1993; *Suivi de l'invasion de l'algue tropicale Caulerpa taxifolia devant en Mediterranée; Situation au 31.12.1992*; Rapport Laboratoire Environnement
marin littoral, Université de Nice-Sophia Antipolis. GIS Posidonie, Marseilles, France, 80 pp;

[3.26] Chisholm J.R.M., Jaubert J.M., 1997; *Photoautotrophic metabolism of Caulerpa taxifolia (Chlorophyta) in the NW Mediterranean*; Mar. Ecol. Prog. Ser. 153:113:123;
[3.27] Komatsu T., Meinesz A., Buckles D., 1997; *Temperature and light responses of alga Caulerpa taxifoliaintroduced into the Mediterranean Sea*; Mar. Ecol. Prog. Ser. 146:145-153;
[3.28] DeVillele X., Verlaque M., 1995; *Changes and degradation in a Posidonia oceanica bed invaded by theintroduced tropical alga Caulerpa taxifolia in the north western Mediterranean*; Bot. Mar. 38:79-87;
[3.29] Schramm W., Booth W., 1981; *Mass bloom of the agla Cladophora prolifera in Bermuda: productivity and phosphorus accumulation*; Bot. Mar. 24:419-426;
[3.30] Gnassia-Barelli M., Lemée R., Pesando D., Romeo M., 1995; *Heavy Metal Distribution in Caulerpa taxifolia from the North-Western Mediterranean*; Marine Pollution Bulletin, Vol.30;
[3.31] Chisholm J.R.M., Dauga C., Ageron E., Grimont P.A.D., Jaubert J.M., 1996; *Roots in mixotrophic algae*; Nature 381:382;
[3.32] Chisholm J.R.M., Moulin P., 2003; *Stimulation of nitrogen fixation in refractory organic sediments by Caulerpa taxifolia (Chlorophyta)*; Limnol. Oceanogr. 48(2):787-794;
[3.33] LaPointe B.E., Toasko D.A., Matzie W.R., 1994; *Eutrophication and trophic state classification of seagrass communities in the Florida Keys*; Bull. Mar. Sci. 54:696-717.
[3.34] Panayotidis P., Montesanto B., 1994; *Caulerpa racemosa (Chlorophyta) on the Greek Coasts*; Cryptogam. Algol. 15:159-161.
[3.35] Doumenge F. 1995; *Quelques re´flexions sur les algues Caulerpes*; Biol. Mar. Mediterr. 2:613-633.
[3.36] Davis A.R., Roberts D.E., Cummins S.P., 1997; *Rapid invasion of a sponge-dominated deep-reef by Caulerpa scalpelliformis (Chlorophyta) in Botany Bay, New South Wales*; Aust. J. Ecol. 22:146-150.
[3.37] Verlaque M., Fritayre P., 1994; *Mediterranean algal communities are changing in the face of the invasive alga Caulerpa taxifolia (Vahl) C. Agardh*; Oceanol. Acta 17:659-672;
[3.38] DeVilléle X. Verlaque M., 1995; *Changes and degradation in a Posidonia oceanica bed invaded by the introduced tropical alga Caulerpa taxifolia in the northwestern Mediterranean*; Bot. Mar. 38:79-87;
[3.39] Garcia E., Rodríguez-Prieto C., Delgado O., Ballesteros E., 1996; *Seasonal light and temperature responses of Caulerpa taxifolia from the north western Mediterranean*; Aquatic Botany, Vol.53:215-225;
[3.40] Ott J.A., 1980; *Growth and Production in Posidonia oceanica (L.) Delile*; Mar. Ecol. 1:47-64;
[3.41] Bay D., 1984; *A field study of the growth dynamics and productivity of Posidonia oceanica (L.) Delile in Calvi Bay, Corsica*; Aquat. Bot. 20:43-64;
[3.42] Ballesteros E., 1989; *Production of seaweed in the north-eastern Mediterranean marine communities: its relation with environmental factors*; Sci.Mar. 53:357-364;
[3.43] Ballesteros E., 1992; *Els vegetals i la zonaciò litoral: espécies, comunitats i factors que influeixen en la seva distribuciò*; Arx.Secc.Ciénc.Inst.Est.Cat. 101:-616
[3.44] Ceccherelli G., Cinelli F., 1997; *Short-term effects of nutrient enrichment of the sediment and interactions between the seagrass Cymodocea nodosa and the introduced green alga Caulerpa taxifolia in a Mediterranean bay*; J.o.Exp.Mar.Biol. Ecol. 217:165-177;

PART-IV

[4.1] Harmelin J.G., 1996; *Des habitats artificiels pour réhabiliter les fonds colonisés par Caulerpa taxifolia*; In: "2nd International Workshop on Caulerpa taxifolia"; Ribera M.A., Ballesteros E., Boudouresque C.F., Gomez A., Gravez V., (Eds.), pp.163-168; University of Barcelona – SPA;
[4.2] Harmelin-Vivien M., Francour P., Harmelin J.G., 1999; *Impact of Caulerpa taxifolia on Mediterranean fish assemblages: a six year study*; In: Proceedings of the Workshop on Invasive Caulerpa in the Mediterranean; MAP Technical Reports Series 125:127-138. Athens: UNEP;
[4.3] Bellan-Santini D., Arnaud P., Bellan G., Verlaque M., 1996; *The influence of the introduced tropical alga Caulerpa taxifolia, on the biodiversity of the Mediterranean marine biota*; Journal of the Marine Biological Association of the UK 76:235-237.
[4.4] Francour P., Harmelin-Vivien M., Harmelin J.G., Duclerc J., 1995; *Impact of Caulerpa taxifolia colonization on the ichthyofauna of North-Western Mediterranean sea: preliminary results*; Hydrobiologia 300/301:345-353;
[4.5] Relini G., Relini M., Torchia G., 2000; *The role of fishing gear in the spreading of allochthonous species: the of Caulerpa taxifolia in the Ligurian Sea*; ICES J.o.Marine Science; Vol.57:1421-1427;
[4.6] Spanier E., Finkeltein V., Raikhlin-Eisehraft B., 1989; *Toxicity of the saupe, Sarpu salpu (Linnaeus, 1758), on the Mediterranean coast of Israel*; J. Fish. Biol., 37:503-504.

[4.7a] Whitehead P.J.P., Bauchot M.L., Hureau J.C., Nielsen, J., Tortonese E., 1986; *Fishes of the North-Eastern Atlantic and the Mediterranean*;Vol. II. Unesco, pp.905
[4.7b] Troell M., Pihl L., Rönnbäck P., Wennhage H., Söderqvist T., Kautsky N., 2004; *When Resilience is Undesirable: Regime Shifts and Ecosystem Service Generation in Swedish Coastal Soft Bottom Habitats*;The Royal Swedish Academy of Sciences; (http://www.beijer.kva.se/publications/pdf-archive/Disc187.pdf);
[4.8] Boudouresque C.F., Meinesz A., 1982; *Découverte de l'herbier de posidonie*; Parc Nat. Port-Cros / Parc Nat. Rég. Corse & GIS Posidonie Edit. Marseille
[4.9] Ott J.A., 1980; *Growth and Production in Posidonia oceanica (L.) Delile*; Mar. Ecol. 1:47-64;
[4.10] Meinesz A., 2000; personal comunication aired via BBC-2, 2001; *Killer Algae*; (http://www.bbc.co.uk/science/horizon/2000/killeralgae.shtml);
[4.11] Agostini S., Desjobert J.M., Pergent G., 1998; *Distribution of phenolic compounds in the seagrass Posidonia oceanica*; Phytochemistry, Vol.48:611-617;
[4.12] deVillele X., Verlaque M., 1994; *Incidence de l'algue introduite Caulerpa taxifolia sur le phytobenthos de Méditerranée occidentale: 1. L'herbier de Posidonia oceanica (L.) Delile*; In: Boudouresque C.F., Meinesz A., Gravez V., (Eds.), "First international workshop on Caulerpa taxifolia"; GIS Posidonie, Marseille: 343-347.
[4.13] Ceccherelli G., Cinelli F., 1997; *Short-term effects of nutrient enrichment of the sediment and interactions between the seagrass Cymodocea nodosa and the introduced green alga Caulerpa taxifolia in a Mediterranean bay*; J.o.Exp. Mar. Biol. Ecol. 217:165-177.
[4.14] Boudouresque C. F., Meinesz A., Ribera M. A., Ballesteros E., 1995; *Spread of the green alga Caulerpa taxifolia (Caulerpales, Chlorophyta) in the Mediterranean: possible consequences of a major ecological event*; Sci.Mar.59 (Suppl.1):21-29;
[4.15] deVilléle X., Verlaque M., 1995; *Changes and degradation in a Posidonia oceanica bed invaded by the introduced tropical alga Caulerpa taxifolia in the north western Mediterranean*; Bot.Mar. 38:79-87;
[4.16] Meinesz A, deVaugelas J., Benichou L., Caye C., Cottalorda J.M., Delehaye L., Febvre M., Garcin S., Komatsu T., Lemée R., Mari X., Molenaar H., Perney L., Venturini A., 1993; *Suivi de l'invasion de l'algue tropicale Caulerpa taxifolia devant en Mediterranée; Situation au 31.12.1992*; Rapport Laboratoire Environnement
marin littoral, Université de Nice-Sophia Antipolis. GIS Posidonie, Marseilles, France, 80 pp;
[4.17] Meinesz A., Benichou L., Blachier J., Komatsu T., Lemée R., Molenaar H., Mari X., 1995; *Variations in the structure, morphology and biomass of Caulerpa taxifolia in the Mediterranean Sea*; Bot.Mar. 38:499-508;
[4.17a] Hofrichter R., 2001; *Das Mittelmeer* Band II/1; Spektrum Akademischer Verlag, Heidelberg – FRG
[4.18] Cuny P., Serve L., Jupin H., Boudouresque C.F., 1995; *Water soluble phenolic compounds of the marine phanerogam Posidonia oceanica in a Mediterranean area colonised by the introduced chlorophyte Caulerpa taxifolia*; Aquatic Botany, Vol.52:237-242;
[4.19] Ceccherelli G., Cinelli F., 1999; *Effects of Posidonia oceanica canopy on Caulerpa taxifolia size in a north-western Mediterranean bay*; Journal of Experimental Marine Biology and Ecology p.19-36;
[4.20] Tippets K., 2002; *The Effects of Caulerpa taxifolia on Invertebrate Abundance in Agua Hedionda Lagoon, California*; (http://ist-socrates.berkeley.edu/~es196/projects/2002final/Tippets.pdf);
[4.22] Ceccherelli G., Cinelli F., 1998: *Habitat effect on spatio-temporal variability of size and density of the introduced alga Caulerpa taxifolia*; Mar. Ecol. Progr. Ser. 163:289-294;
[4.23] Verlaque M., Fritayre P., 1994; *Modifications des communautés algales méditerranéennes en présence de l'algue envahissante Caulerpa taxifolia (Vahl) C.Agardh*; Oceanol. Acta 17:659-672;
[4.24] Ferrer E., Garreta A.G., Ribera M.A., 1997; *Effect of Caulerpa taxifolia on the productivity of two Mediterranean macrophytes*; Mar. Ecol. Prog. Ser. 149:279-287;
[4.25] Belsher T., Meinesz A., 1995; *Deep-water dispersal of the tropical alga Caulerpa taxifolia introduced into the Mediterranean*; Aquatic Botany, Vol.51:163-169;
[4.26] Bellan-Santini D., Arnaud P.M., Bellan G., Verlaque M., 1996; *The influence of the introduced tropical alga Caulerpa taxifolia, on the biodiversity of the Mediterranean marine biota*; J.o.Mar.Biol. Assoc. U.K. 76:235-237.
[4.27] Pedrotti M.L., Lemée R., 1999; *Effect of microalgae treated with natural toxins on the nutrition and development of filter-feeding sea-urchin larvae*; Marine Environmental Research Vol.48:177-192;
[4.28] Boudouresque C.F., Lemée R., Mari X., Meinesz A., 1996; *The invasive alga Caulerpa taxifolia is not a suitable diet for the sea urchin Paracentrotus lividus*; Aquatic Botany; Vol.53:245-250;

[4.29] McConnel O.J., Hughes P.A., Targett N.M., Daley J., 1982; *Effects of secondary metabolites from marine algae on feeding by the seaurchin Lytechinus variegatus*; J.o.Chem. Ecol. 8:1437-1453;
[4.30] Amade P., Lemée R., 1998; *Chemical defence of the mediterranean alga Caulerpa taxifolia variations in caulerpenyne production*; Aquatic Toxicology, Vol.43:287-300;
[4.30a] Relini G., Relini M., Torchia G., 2000; *Fish population changes following the invasion of the allochtonous alga C.taxifolia in the Ligurian Sea (N-W Mediterranean)*; International Council for the Exploration of the Sea;
[4.31] Endler J.A., 1978; *A predator's view of animal colour patterns*; In: Evolutionary Biology 11 (Steere, W. C. & Wallace, B., eds.), pp.319-364. New York: Plenum Press.
[4.31] Crook A.C., 1997; *Colour patterns in coral reef fish. Is background complexity important?*; Journal of Experimental Marine Biology and Ecology 217:237-252;
[4.32] Planes S., Doherty P.J., 1997; *Genetic and colour interactions at contact zone of Acanthochromis polyacanthus: a marine fish lacking pelagic larvae*; Evolution 51:1232-1243;
[4.33] Francour A.P., Harmelin-Vivien M., Zaninetti L., 2002; *Adaptive colouration of Mediterranean labrid fishes to the new habitat provided by the introduced tropical alga Caulerpa taxifolia*; Journal of Fish Biology, Vol.60:1486-1497;
[4.34] Francour P., Harmelin-Vivien M., Harmelin J.G., Duclerc J., 1995; *Impact of Caulerpa taxifolia colonization on the ichthyofauna of North-Western Mediterranean sea: preliminary results*; Hydrobiologia 300/301:345-353;
[4.35] Merilaita S., Tuomi J., Jormalainen V., 1999; *Optimization of cryptic coloration in heterogeneous habitats*; Biological Journal of the Linnean Society 67:151-161;
[4.36] Paul V.J. and Fenical W., 1986; *Chemical defense in tropical green algae, order Caulerpales*; Mar.Ecol.Prog.Ser., 34:157-169.
[4.37] Faulkner D.J., 1987; *Marine natural products*; Nat. Prod. Rep., 1:540-542;
[4.38] Schwede J.G., Cardellina J.H., Grode S.H., James T.R., Blackmann A.J., 1987; *Distribution of the pigment caulerpin in species of the green alga Caulerpa*; Phytochemistry, 20:155-158;
[4.39] Paul V.J. 2002; *Chemical Ecology of Caulerpa spp. with an Emphasis on Invasive Caulerpa taxifolia*: University o.Guam Marine Lab. - USA; (http://sgnis.org/update/ctax.htm);
[4.40] Ayyad S.E.N., Badria F.A., 1994; *Caulerpin, an antitumor indole alkaloid from Caulerpa racemosa*; Alex. J. Pharm. Sci. 8:217.
[4.41] Pesando D., Lemée R., Ferrua C., Amade P., Girard J.P., 1996; *Effects of caulerpenyne, the major toxin from Caulerpa taxifolia on mechanisms related to sea urchin egg cleavage*; Aquatic Toxicology, Vol.35:139-155;
[4.42] Hodgson L.M., 1984; *Antimicrobial and antineoplasic activity in some South Florida seaweeds*; Bot. Mar. 27: 387-390;
[4.43] Lemée R., Pesando D., Durand-Clament M., Dubreuil A., Meinesz A., Guerriero A., Pietra F., 1993; *Preliminary survey of toxicity of the green alga Caulerpa taxifolia introduced into the Mediterranean*;J.o.Appl. Phycol. 5:485-493;
[4.44] Pesando D., Lemée R., Durand-Clement M., Dubreuil A., Amade P., Valls R., Berhaud J., Pedrotti M.L., Fenaux L., Guerriero A., Pietra F., Meinesz A., 1994; *Risques liés a la toxicité de Caulerpa taxifolia (Vahl) C. Agardh*; In: Boudouresque C.F., Meinesz A., Gravez V.. (Eds.), "First International Workshop on Caulerpa taxifolia"; GIS Posidonie, Marseille, France, pp. 265-269;
[4.45] Lemée R., Pesando D., Issanchou C., Amade P., 1997; *Microalgae: a Model to Investigate the Ecotoxicity of the Green Alga Caulerpa taxifolia from the Mediterranean Sea*; Marine Environmental Research; Vol.44:13-25;
[4.46] Guerriero A., Meinesz A., D'Ambrosio M., Pietra F., 1992; *Isolation of toxic and potentially toxic sesqui- and monoterpenes from the tropical green seaweed Caulerpa taxifolia which has invaded the region of Cap Martin and Monaco*; Helv. Chim. Acta 75:689-695;
[4.47] Dumay O., Pergent G., Pergent-Martini C., Amade P., 2002 *Variations in Caulerpenyne contents in Caulerpa taxifolia and .racemosa*; J.o.Chemical Ecology Vol.28;
[4.48] Guerriero A., D'Ambrosio M., Guella G., Dini F., Pietra F., 1994; *Secondary metabolites of the green seaweed Caulerpa taxifolia introduced into the Mediterranean sea, and a comparison with ciliates of the genus Euplotes*; In: Boudouresque C.F., Meinesz A., Gravez V., (Eds.); "First International Workshop on Caulerpa taxifolia"; GIS Posidonie Publication.
[4.49] Jung V., Pohnert G., 2001; *Rapid wound-activated transformation of the green algal defensive metabolite*

caulerpenyne; Tetrahedron, Vol.57:7169-7172;
4.50 Parrain J.L., Santelli M., 2002; *Totale Synthesis of Terpenoids Isolated from Caulerpa*; Antibes - Juan les Pins, Palais des Congrès; (http://www.unice.fr/cdiec/congres/sfc_paca/sfc_2002_italienne.htm);
4.51 Amade P., Lemée R., 1998; *Chemical defence of the mediterranean alga Caulerpa taxifolia variations in caulerpenyne production*; Aquatic Toxicology, Vol.43:287-300;
4.52 Dumay O., Pergent G., Pergent-Martini C., Amade P., 2002 *Variations in Caulerpenyne contents in Caulerpa taxifolia and C.racemosa*; J.o.Chemical Ecology Vol.28;
4.53 Amade P., Valls R., Bouaicha N., Lemée R., Artraud J. 1994; *Méthodes de dosage de la caulerpényne produite par Caulerpa taxifolia*; pp. 163-167: In Boudouresque C.F., Meinesz A., Gravez V., (Eds.); "First International Workshop on Caulerpa taxifolia"; GIS Posidonie.
4.54 Meyer K.D., Paul V.J., 1992; *Intraplant variation in secondary metabolite concentration in three species of Caulerpa (Chlorophyta: Caulerpales) and its effects on herbivorous fishes*; Mar. Ecol. Prog. Ser. 82:249-257;
4.55 Giannotti A., Ghelardi E., Senesi S., 1994; *Characterization of seawater bacterial communities within environments colonized by the tropical green seaweed Caulerpa taxifolia*; In: Boudouresque C.F., Meinesz A., Gravez V. (Eds.), "First International Workshop on Caulerpa taxifolia"; GIS Posidonie, Marseille, pp. 197-201;
4.56 Dini F., Rosati G., Erra F., Verni F., Pietra F., 1994: *The environmental toxicity of secondary metabolites produced by the Mediterranean -adapted seaweed Caulerpa taxifolia using marine ciliate protists as a model*; In: Boudouresque C.F., Meinesz A., Gravez V. (Eds.), "First International Workshop on Caulerpa taxifolia"; GIS Posidonie, Marseille, pp. 355-364;
4.57 Barbier P., Guise S., Huitorel P., Amade P., Pesando D., Briand C., Peyrot V., 2001; *Caulerpenyne from Caulerpa taxifolia has an antiproliferative activity on tumor cell line SK-N-SH and modifies the microtubule network*; Life Sciences, Vol.70:415-429;
4.58 Fischel J.L., Lemée R., Formento P., Caldani C., Moll J.L., Pesando D., Meinesz A., Grelier P., Pietra F., Guerriero A., Milano G., 1995; *Cell growth inhibitory effects of caulerpenyne, a sesquiterpenoid from the marine alga Caulerpa taxifolia*;. Anticancer Res. 15:2155-2160:
4.59 DeHaro L., Treffot M.J., Jouglard J., Perringue C., 1993; *Trois cas d'intoxication de type ciguateresque aprés ingestion de sparidae de Mediterranée*; Ictyol. Physiol. Acta 16:133-146;
4.60 Brunelli M., Garcia-Gil M., Mozzachiodi R., Roberto M., Scuri R., Traina G., Zaccardi M.L., 2000; *Neurotoxic Effects of Caulerpenyne*; Progress in Neuro-Psychopharmacology & Biological Psychiatry, Vol.24:939-954;
4.61 Schröder H.C., Badria F.A., Ayyad S.N., Batel R., Wiens M., Hassanein H.M.A., Kurelec B., Müller W.E.G., 1998; *Inhibitory effects of extracts from the marine alga Caulerpa taxifolia and of toxin from Caulerpa racemosa on multixenobiotic resistance in the marine sponge Geodia cydonium*; Environ.Tox. & Pharm. Vol.5:119-126;

PART-V

5.1 Paul V.J., Hay M.E., 1986; *Seaweed susceptibility to herbivory: chemical and morphological correlates*; Marine Ecology Press Series 33:255-264;
5.2 Jensen K.R., 1994; *Behavioural adaptations and diet specificity of Sacoglossan Opisthobranchs*; Ethol. Ecol. and Evol. 6:87-101;
5.3 Thibaut T., Meniesz A., 2000; *Are the Mediterranean scoglossan molluscs Oxynoe olivacea and Lobiger serradifalci suitable agents for a biological control against the invading alga Caulerpa taxifolia*; Comples Rendu de l'Académie des Sciences, Paris, Life Sciences, 323:477-488;
5.3a Meinesz A., Melnick J., Blachier J., Charrier S.; 1996; *Etude préliminaire,en aquarium, de deux ascoglosses tropicaux concommantCaulerpa taxifolia: une voie de recherce pour la lutte biologique*; in Proceedings of the 2nd International workshop on Caulerpa taxifolia, Barcelona – ESP, Ribera et al (ed);Universidad Barcelona Publications; pp.157-161;
5.4 Clark K.B., Busacca M., Stirts H., 1979; *Nutritional aspects of development of the ascoglossan, Elysia cauze*; In: Stancyk (Ed.); Reproductive ecology of marine invertebrates pp.11-24; Columbia University of South Carolina - USA;
5.5 Clark K.B., Jensen K.R., 1981; *A comparison of egg size, capsule size and develolment patterns in the order Ascoglossa (Sacoglossa) (Mollusca: Opisthobranchia)*; International Journal of Invertebrate Reproduction 3:57-64;

[5.6] Thibaut T., Meinesz A., Amade P., Charrier S., DeAngelis K., Ierardi S., Mangialajo L., Melnick J., Vidal V., 2001; *Elysia subornata (Mollusca) a potential control agent of the alga Caulerpa taxifolia (Chlorophyta) in the Mediterranean Sea*; Journal of Marine Biol. Ass.;
[5.7] Clark, K.B., Jensen, K.R., Stirts, M.H., 1990; *Survey for functionnal kleptoplasty among West Atlantic Ascoglossa (Sacoglossa) (Mollusca: Opisthobranchia)*; The Veliger 33:259-280.
[5.8] Meinesz A., Benichou L., Blachier J., Komatsu T., Lemée R., Molenaar H., Mari X., 1995; *Variations in the structure, morphology and biomass of Caulerpa taxifolia in the Mediterranean Sea*; Bot. Mar. 38, 499-508.
[5.9] Coquillard P., Thibaut T., Hill D.R.C., Gueugnot J., Mazel C., Coquillard Y., 2000; *Simulation of the mollusc Ascoglossa Elysia subornata population dynamics: application to the potential biocontrol of Caulerpataxifolia growth in the Mediterranean Sea*; Ecological Modelling, Vol.135:1-16;
[5.10] Food and Agriculture Organization of the United Nations (FAO), 1997; *Code of conduct for the import and release of exotic biological control agents*; Biocontrol News Information 18:118N-124N; (http://www.fao.org/docrep/x5585E/x5585e0i.htm);
[5.11] International Council for the Exploitation of the Seas (ICES), 1997; *Study group on marine biocontrol of invasive species*; La Tremblade 1997, Copenhagen - DK; (http://www.ices.dk/);
[5.12] Pierce S.K., Maugel T.K. Rumpho M.E., Hanten J.J., Mondy W.L., 1999; *Annual viral expression in a sea slug population: life cycle control and symbiotic chloroplast maintenance*; Biological Bulletin, Marine Biological Lab. Woods Hole 197:1-6;
[5.13] Zuljevic, A., Antolic B., 2002; *Reproduction of Caulerpa taxifolia in the Mediterranean Sea*; International Caulerpa taxifolia Conference, Institute of Oceanography and Fisheries; (http://sgnis.org/update/ctax.htm);
[5.13a] Creese R.G., Davis A.R. Glasby T.M.; 2004; *Eradicating and preventing the spread of the invasive alga Caulerpa taxifolia in NSW*; NSW Fisheries Final Report Series No.: 64;NSW Fisheries, Cronulla – AUS
[5.14] NZ Fisheries and Marine Biosecurity (NZFMB), 2001; *Action Plan for unwanted species*; Ministery of Fisheries, Wellington - NZ (http://www.biodiversity.govt.nz/pdfs/seas/c_taxifolia_action_plan.pdf);
[5.15] Relini G., Relini M., Torchia G., 2000; *The role of fishing gear in the spreading of allochthonous species: the of Caulerpa taxifolia in the Ligurian Sea*; ICES Journal of Marine Science; Vol.57;1421-1427;
[5.16] Thibaut, T., Meinesz A., 2002; *Management Successes and Failures in the Mediterranean*; Université de Nice-Sophia Antipolis - FRA (http://sgnis.org/update/ctax.htm);
[5.17] Riera F., Pou S., Grau A.M., Delgado O., Weitzmann B., Ballesteros E., 1994; *Eradication of a population of the tropical green alga Caulerpa taxifolia in Cala d'Or (Mallorca, Western Mediterranean): methods and results*; In: Boudouresque C.F., Meinesz A., Gravez V., (Eds.); "First International Workshop on Caulerpa taxifolia"; GIS Posidonie Publ, France, pp. 327-331;
[5.18] Millar A., Talbot B., 2002; *The Introduction of Caulerpa taxifolia in New South Wales, Australia*; Royal Botanic Gardens Sydney - AUS; (http://sgnis.org/update/ctax.htm);
[5.19] Withgott J., 2002; *California Tries to Rub Out the Monster of the Lagoon*; Science 295:2201-2202;
[5.20] Merkel K., Woodfield R., 2003*Eradication and Surveillance of Caulerpa taxifolia within Agua Hedionda Lagoon, Carlsbad, California: Second Year Status Report Fall 2001 through Summer 2002*;Merkel & Associates, Inc. (CA) - USA; (http://www.swrcb.ca.gov/rwqcb9/programs/caulerpa/caulerpa.html);
[5.21] Hugues R.H., 1980; *Optimal foraging theory in the marine context*; Oceanographic and Marine Biology, Annual Review 18:423-481;
[5.22] Carlton JT., 1997; *La lutte biologique, avantages et risques*; In: Séminaire international "dynamique d'espéces marines invasives: application al'expansion de Caulerpa taxifolia en Mediterranée", Académie des Sciences, Paris, 13-15.03.1997. Actes de Colloques (ed. Académie des Sciences), pp.279-284. Paris, Lavoisier Publishing;
[5.23] Vaugelas J., Meinesz A., Antolic B., Ballesteros E., Belsher T., Cassar N., Ceccherelli G., Cinelli F., Cottalorda J.M., Orestano C.F., 1999; *Standardization proposal for the mapping of Caulerpa taxifolia expansion in the Mediterranean Sea*; Oceanologica Acta, Vol.22:85-94;
[5.24] Aussem A., Hill D., 1999; *Wedding connectionist and algorithmic modelling towards forecasting Caulerpa taxifolia development in the north-western Mediterranean sea*; Ecological Modelling, Vol.120:225-236;
[5.25] Lemée R., Pesando D., Issanchou C., Amade P., 1997; *Microalgae: a Model to Investigate the Ecotoxicity of the Green Alga Caulerpa taxifolia from the Mediterranean Sea*; Marine Environmental Research; Vol.44:13-25;
[5.26] Ribera M.A., Boudouresque C.F., 1995; *Introduced marine plants, with special reference to macroalgae: mechanisms and impact*; In: Round F.E., Chapman D.J. (Eds.); Progress in Phycological Research, Vol. 11:187-268;

[5.27] Meinesz A., 1992; *Modes de dissémination de l'algue Caulerpa taxifolia en Méditerranée*; Rap. Comm. internation. Explor. sci. Médit., 33:B44;
[5.28] Sant N., Delegado O., Roddriguez-Prietro C., Ballesteros E., 1996; *The spreading of the introduced seaweed Caulerpa taxifolia (Vahl) C. Agardh in the Mediterranean sea: testing the boat transportation hypothesys*; Botanica Marina, 39:427-430;
[5.29] Boudouresque C.F., Verlaque M., 2002; *Biological pollution in the Mediterranean Sea: invasive versus introduced macrophytes*; Marine Pollution Bulletin, Vol.44:32-38;
[5.31] Schaffelke B., Murphy N.; Uthicke S., 2002; *Using genetic techniques to the sources of the invasive alga Caulerpa taxifolia in three new locations in Australia*; Marine Pollution Bulletin, Vol.44:204-210;
[5.32] Johnson S.C., 2001; *Invasion of "Killer" Mediterranean Weed to California, USA*; (http://www.crc.uri.edu/comm/download/IC_40/Johnson.pdf);
[5.33] Department for Environment Food and Rural Affairs (DEFRA), 2003; *Review of non-native species policy; Report of the working group*; London – UK; (http://www.defra.gov.uk/wildlife-countryside/resprog/findings/non-native/report.pdf);
[5.34] McClary D., Nelligan R.J., 2001; *Alternate Biosecurity Management Tools for Vector Threats - Technical Guidelines for Acceptable Hull Cleaning Facilities*; New Zealand Ministery of Fisheries (http://www.fish.govt.nz/sustainability/biosecurity/research/zbs2000_03-kma.pdf);
[5.35] Allendorf F.W., Lundquist (2003; *Population Biology of Invasive Species*; Conservation Biology, Pages 24-30, Volume 17, No. 1; (http://www.blackwell-synergy.com/links/doi/10.1046/j.1523-1739.2003.02365.x/full/);

PART-VI
[6.0]ISSG, 2004; Invasive Species Specialist Group of the World Conservation Union (IUCN) (http://www.issg.org/database/species/search.asp?st=100ss&fr=1&sts=#);
[6.1] Ceccherelli G., Cinelli F., 1997; *Short-term effects of nutrient enrichment of the sediment and interactions between the seagrass Cymodocea nodosa and the introduced green alga Caulerpa taxifolia in a Mediterranean bay*; J.o.Exp.Mar.Biol. Ecol. 217:165-177;
[6.2] Piazzi L., Cinelli F., 2000; *Effects of the spread of the introduced Rhodophyceae Acrothamnion preissii and Womersleyella setacea on the macroalgal community of Posidonia oceanica rhizomes in the Western Mediterranean sea*; Cryptogam., Algol. 21(3):291-300;
[6.3] Rejmánek M., 1989; *Invasibility of plant communities*; In: Drake J.A., Mooney H.A., DiCastri F., Groves R.H., Kruger F.J., Rejmánek M., Williamson M. (Eds.); Biological Invasions: a Global Perspective. Wiley, Chichester, pp. 369-388;
[6.4] Case T.J., 1990; *Invasion resistance arises in strongly interacting species-rich model competition communities*; Proc. Natl. Acad. Sci. USA 87:9610-9614;
[6.5] Law R., Morton D., 1996; *Permanence and the assembly of ecological communities*; Ecology 77:762-775;
[6.6] Stachowicz J.J., Whitlatch R.B., Osman R.W., 1999; *Species diversity and invasion resistance in a marine ecosystem*; Science 286:1577-1579;
[6.7] Prieur-Richard A.H., Lavorel S., 2000; *Invasions: the perspective of diverse plant communities*; Austral Ecol. 25:1-7;
[6.8] Carlton J.T., Geller J.B., 1993; *Ecological roulette: the global transport of nonindigenous marine organisms*; Science 261:78-82;