# Extension of two Caulerpa species along the Tunisian coast

Langar, H.1\*; Djellouli, A.S.2; Sellem, F.1 & El Abed, A.1

<sup>1</sup>Intitut National des Sciences et Technologies de la Mer (I.N.S.T.M), 28, Rue du 2 mars 1934, 2025 Salammbô, Tunisia; <sup>2</sup>Faculté des Sciences de Tunis, Campus universitaire, 1060 Tunis, Tunisia; \*Corresponding author; Fax +21671732622; E-mail habib.langar@instm.rnrt.tn

Abstract. Two species of the green alga genus Caulerpa are considered as invasive in Tunisia: Caulerpa racemosa and C. taxifolia. C. racemosa is found in a many new localities at the central and southern Tunisian coast, and even in the North of the country in colder water. These new observations lead to the hypothesis of the multiplicity of the origins of the introduction of the species and suggest possible invasive potentialities. The general thallus, and more particularly the number, the shape as well as the arrangement of ramelli on fronds allowed us to identify four morphologically different groups. These groups could be either different systematic entities or polymorphic variations of the same taxon due to different ecological conditions. Caulerpa taxifolia was observed for the first time in Tunisia at the roadstead of Sousse in the beginning of the year 2000. It is recognized as identical to that found in the other Mediterranean localities (GenBank number AJ228960). Up to the end of March 2000, a submarine prospecting campaign revealed an affected area of about 350 ha in front of 2 km of coast. Colonies of Caulerpa taxifolia disseminating in this area appear in isolated spots. form, In the growing stage, they form surfaces varying between 0.5 to 6 m<sup>2</sup> and located 20 to 100 m from each other. The total covered surface was estimated to be between 0.5 and 1 ha.

**Keywords:** Caulerpa racemosa; Caulerpa taxifolia; Distribution; Invasive species.

### Introduction

Caulerpales are generally considered as a tropical and subtropical order of green algae which is also found in warm-temperate waters. Among the species of the genus Caulerpa inventoried in the Mediterranean, two are of special interest in Tunisia: Caulerpa racemosa (Forsskål) J. Agardh, and Caulerpa taxifolia (Vahl) C. Agardh. C. racemosa was reported for Tunisia by Hamel (1926) in the first half of the 20th century. C. taxifolia, since 1984 present on the northwestern banks of the Mediterranean Sea (Meinesz & Hesse 1991), was reported for the south banks only in Tunisia (Langar et al. 2000). The many reports on C. taxifolia in the northwestern Mediterranean, the recent new findingss of C. racemosa throughout the Mediterranean littoral and the qualification of these species as invasive, have triggered monitoring programs in all countries bordering the Mediterranean Sea (Meinesz et al. 2001). The present study was carried out within the framework of the control of appearance of new marine species (introduction or migration) and of the prevention of their expansion.

## **Material and Methods**

During diving excursions arranged regularly along the Tunisian coast, several new localities of *Caulerpa racemosa* were recently discovered accidentally. These localities were listed and are since then the object of a regular follow-up. From each locality, a sample of *C. racemosa* was collected and preserved either in a solution of formaldehyde/sea water (1:19) or simply in the refrigerator until further analysis. Furthermore, fragments of thallus of *C. racemosa* have been brought by fishermen or amateur divers from other localities which we did not survey. In view of the bad conservation status of these fragments, they were not used for the descriptive study, but they have been used as indicators for a refinement of the geographical distribution of the species along the Tunisian coast.

The discovery of C. taxifolia in marine Tunisian

waters in the beginning of the year 2000 (Langar et al. 2000) was the result of a prevention and awareness campaign launched in 1997 (Langar et al. 1998). The cartography of the zone where the presence of this seaweed was established, has been achieved at the end of March 2000, following an underwater prospecting campaign in the zone where occurrence was suspected. This was realized as follows. In the latter zone, ten well trained divers were successively towed from the surface following pre-defined transects. Each time a given diver encountered a colony of the algae, the place was marked using a floating signal and a brief description of the colony was made. At surface, the position of each floating signal was reported on a map, using a GPS, and the perimeter of the zone colonized by C. taxifolia was inserted on the map.

A genetic identification of the strain of *C. taxifolia* found in Tunisia, was made according to Jousson et al. (1998) after DNA extraction, cloning and sequencing.

## **Results and Discussion**

Caulerpa racemosa: current distribution and morphological variations

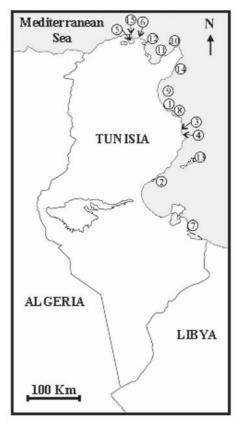
The compilation of observations of Caulerpa racemosa allowed us to evaluate the extension of its current distribution area in Tunisia (Fig. 1): In August 1996, the alga was collected in the Lagoon and the Canal of Bizerte in the north of the country. In the same region, it was observed in August 1997 near Cani island. In 1998, it was brought from Zarzis (south of the Golf of Gabes). In March of the same year it was collected in the bay of Monastir and in September, in the Golf of Hammamet. During the summer of 1999, it was observed at Sidi Daoud, Ras Fartas and Korbous, in the West littoral of the Cap Bon, as well as at Metline and Rafraf, in the North of the Gulf of Tunis. Finally in the course of the summer of 2000 it was found in the harbour of El Ataya at the Kerkennah islands situated in the east-centre of the Tunisian coast, and in April 2001 it was discovered at Beni Khiar in the southern littoral of Cap Bon and in the harbour of Bizerte.

By referring to the aspect of the general thallus and more particularly to the number, the shape and the arrangement of the ramelli on fronds, four morphologically different groups of *Caulerpa racemosa* were distinguished among the collected plants.

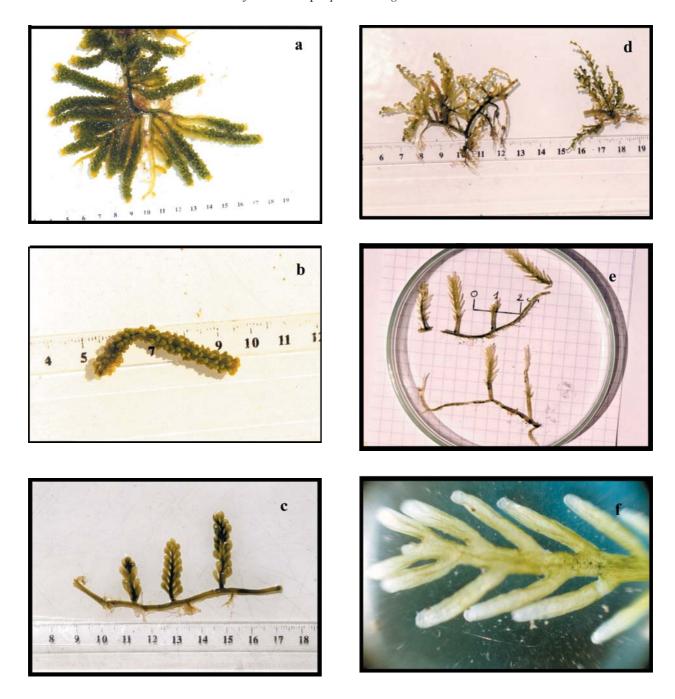
The first grouping is based on samples collected at Monastir, Hammamet, Bizerte and at Cani islands. The thallus (Fig. 2a) forms colonies of ca. 40 cm in diameter. Stolons, of 2 to 3 mm in diameter, are unevenly branched and finely tangled up. They are fixed to the substratum

by groups of rhizoids irregularly arranged. Chlorophyllous in the base, these rhizoids become quickly colourless. Fronds (Fig. 2b) are narrowly spaced from each other. The distance separating their insertion point on the stolon is less than 1 cm. Their mean length varies from 2 to 8 cm and they bear stem-like vesicular ramelli arranged like a row of eight leaves. The mean length of these ramelli and their mean diameter in their middle are respectively ca. 3.8 mm and 800 (600-950)  $\mu m$ . In the top, the ramelli have a flattened or even convex shape, where their diameter can reach 2200 (1900- 2600)  $\mu m$ .

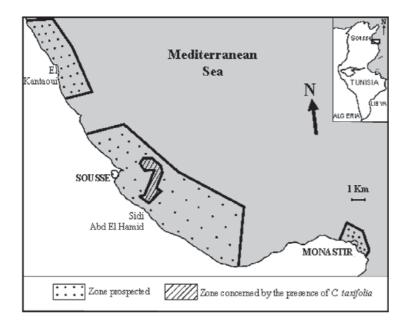
The second grouping corresponds to specimens reported at Cap Bon. The thallus (Fig. 2c) forms colonies stretching over 10-30 m<sup>2</sup>. Stolons, with a diameter of 1 to 1.5 mm, are little branched and the fronds are spaced by 2 to 70 mm, with a mean distance of 14 mm. These fronds have a mean length of 15 mm. They bear alternate ramelli, arranged like two to four leaves. The ramelli gradually expand from the base towards the apex in a droplet shape. Their length varies from 4 to 5



**Fig. 1.** Distribution of *Caulerpa racemosa* in Tunisia. 1 = Sousse harbour; 2 = Golf of Gabes; 3 = Mahdia harbour; 4 = Salakta; 5 = Lagoon of Bizerte; 6 = Cani islands 7 = Zarzis; 8 = Monastir; 9 = Golf of Hammamet; 10 = Sidi Daoud; 11 = Korbous and Ras Fartas; 12 = Metline and Rafraf; 13 = Kerkennah islands; 14 = Beni Khiar; 15 = Bizerte.



**Fig. 2.** Various groups of *Caulerpa racemosa* found in the Tunisian littoral. **a, c, d, e.** General aspect of the thallus belonging to the groups 1, 2, 3 and 4, respectively; **b., f.** Close-ups of fronds belonging to groups 1 and 4, respectively. All scales in cm.



**Fig. 3.** Map of the prospected zone and the zone where the presence of *Caulerpa taxifolia* was established by the end of March 2000.

mm and their diameter ca. 1 mm at the base and 2 mm at the top.

Samples collected in Monastir Bay belong to a third group (Fig. 2d). They were found fixed to *Posidonia oceanica* rhizomes where they formed small colonies grouping ca. ten fronds. Fronds are separated by a distance of 2 to 5 mm. These fronds can be branched and their length varies between 25 and 55 mm. They bear in a spaced out way a clavate ramelli arranged like two or three leaves. The diameter of the ramelli at the base is 750 (700-950)  $\mu$ m. The top of the ramelli is flattened with a diameter from 1.8 to 2 mm.

Group 4 includes specimens collected from the harbour at Kerkennah. It is characterized by unbranched stolons, attached with numerous rhizoidal branchlets (Fig. 2e). Fronds, with a length of maximally 8 cm, bear ramelli of 6 mm maximal length, spaced out and generally cylindrical (Fig. 2f) with a mean diameter of 1 mm.

Morphological differences noticed on Tunisian samples of *C. racemosa* suggest a multiplicity of origins, each corresponding to a form or a different variety of the species. However, different distribution areas, each corresponding to a given morphological group also suggest that variation may be due to environmental factors, e.g. light, habitat type and depth. However, while the populations of Sousse seem to be stable since 1926 and occupy only restricted surfaces (< 50 cm²), those recently discovered at Cap Bon belonging to the second group, have a tendency to expand and occupy much larger areas, and thus behave like an invasive species. Consequently, the latter populations need a more detailed investigation in order to better control their development.

Caulerpa taxifolia: origin and distribution

The area prospected during the submarine campaign, realized in March 2000, was ca. 9250 ha. Three zones were involved in this campaign. The first zone with an area of 6900 ha extends at both sides of the roadstead of Sousse parallel to the coast. The second zone more in the northwest, situated in front of the marina of El Kantaoui, has an area of 2000 ha. The third zone occurring localized in the southeast, in front of the marina of Monastir, covers an area of 350 ha (Fig. 3).

Caulerpa taxifolia was only found in the zone of Sousse on an area of 350 ha along 2 km of the coast, from the roadstead at 17 m depth to the front of this zone of Sidi Abd El Hamid to the southeast of Sousse at a depth of 7 m (Fig. 3).

In the roadstead, *C. taxifolia* is sprouting either directly on loose sandy-muddy sediment particularly in furrows left by the chains of anchors of ships, or on the edge of *Posidonia* beds or even inside damaged beds of *Posidonia*. Dead *Posidonia* beds in this roadstead are often eroded at all heights (up to 2 m) and colonies of *C. taxifolia* are often present on the ledge of these beds.

Towards shallower zones and away from the roadstead, *C. taxifolia* is less frequent, and when it is present, it is found on all types of bottom, infiltrating even *Posidonia* beds.

Colonies of *C. taxifolia* disseminated in the delimited zone appear as isolated spots, in a growing stage, with surfaces varying from 0.5 to  $6 \, \text{m}^2$ , 20 to  $100 \, \text{m}$  from each other, with fronds not exceeding 35 cm in length. The total surface covered was estimated  $0.5 - 1 \, \text{ha}$ .

The presence of *C. taxifolia* and its distribution at a

roadstead and its neighbourhood suggest that the species was introduced through the anchorage of trade ships which had anchored before in an infested zone. Consequently, it would be the same strain as the one already existent in the northwestern Mediterranean Sea. This was confirmed by genetic analysis revealing that the three ITS rDNA cloned fragments and sequenced within an individual, showed an identical sequence as the other *C. taxifolia* specimens collected in different Mediterranean localities (GenBank number AJ228960).

The discovery of *C. taxifolia* in Tunisia over an area of 350 ha suggests that the introduction of the alga occurred several years ago, but an exact dating is not possible. Neither can we judge the classification of the zone where the alga is present according to the system proposed by Vaugelas et al. (1999). The invasive character of the species and its presence in new particular environmental conditions (higher temperature than in the northwest Mediterranean) should encourage us to extend and intensify the monitoring of our littoral.

**Acknowledgements.** The authors are particularly grateful to Professor Alexandre Meinesz from the University of Nice, Sophia Antipolis for her cooperation and to Dr. Olivier Jousson, from the Department of Zoology and Animal Biology of the University of Geneva, for genetic analysis.

### References

- Hamel, G. 1926. Quelques algues rares ou nouvelles pour la flore méditerranéenne. Bull. Mus. Natl. Sci. Nat. Paris 32(6): 420.
- 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. *Mar. Ecol. Progr. Ser.*, 172: 275-280.
- Langar, H., Djellouli, A., Ben Mustapha, K. & El Abed, A. 1998. Campagne de prévention et de sensibilisation à l'introduction en Tunisie de Caulerpa taxifolia. In: Boudouresque, C.F., Gravez, V., Meinesz A. & Pally, F. (eds.) Third International Workshop on Caulerpa taxifolia, Marseille, 19-20 septembre 1997, pp. 17-20. GIS Posidonie, FR.
- Langar, H., Djellouli, A., Ben Mustapha, K. & El Abed, A.
  2000. Première signalisation de *Caulerpa taxifolia* (Vahl)
  J. Agardh en Tunisie. *Bull. Inst. Natn. Sci. Tech. Mer* (*Tunisie*), 27(1): 1-8.
- Meinesz, A. & Hesse, B. 1991. Introduction et invasion de l'algue tropicale *Caulerpa taxifolia* en Méditerranée nordoccidentale. *Oceanol. Acta* 14: 415-426.
- Meinesz, A., Belsher, T., Thibaut, T., Antolic, B., Ben Mustapha, K., Boudouresque, C.F., Chiaverini, D., Cinelli, F., Cottalorda, J.-M., Djellouli, A., El Abed, A, Orestano, C., Grau, A.M., Ivesa, L., Jaklin, A., Langar, H., Massuti-Pascual, E., Peirano, A., Tunesi, L., De Vaugelas, J., Zavodnik, N. & Zuljevic, A. 2001. The introduced green alga *Caulerpa taxifolia* continues to spread in the Mediterranean. *Biol. Invasion* 3: 201-210.
- Vaugelas, J., Meinesz, A., Antolic, B., Ballesteros, E., Belsher,
  T., Cassar, N., Ceccherelli, G., Cinelli, F., Cottalorda, J.
  M., Frada'Orestano, C.M., Grau, A., Jaklin, A., Morucci,
  C., Relini, M., Sandulli, R., Span, A., Tripaldi, G., Van
  Klaveren, P., Zavodnik, N. & Zuljevic, A. 1999. Proposition for a standardization of map representation of *Caulerpa*taxifolia expansion in the Mediterranean Sea. *Oceanol*.
  Acta 22: 85-94.

Received 28 May 2002; Accepted 7 December 2002. Final revision received 17 December 2002; Coordinating Editor: Chr. Pergent-Martini.