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## Population structure and conservation status of the white gorgonian *Eunicella singularis* (Esper, 1791) in Tunisian waters (Central Mediterranean)

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### Abstract

The white gorgonian, *Eunicella singularis*, is thriving in Mediterranean hard-bottom communities; however, data regarding its distribution and ecology remain absent and insufficient, particularly in the southern Mediterranean Sea. In this study, the population structure and disturbance levels of the most common gorgonian in Tunisia were assessed for the first time. During two years (2015-2016), a total of 818 colonies of *E. singularis* were surveyed in five coastal sites, by scuba diving, between 7 to 40 m depth. Collected data included density, colony height, and extent of injury. Mean population density was  $11.91 \pm 7.42$  colonies per m<sup>2</sup> (mean  $\pm$  SD). Mean and maximum colony heights were  $16.49 \pm 5.59$  cm and 51 cm, respectively. Among populations, mean extent of tissue injury differed considerably, ranging from 12.47% to 58.88% and most affected colonies showed old necrosis. These data regarding the demographic structure and level of injuries are needed to provide insights into the conservation status of the Tunisian population of *E. singularis*. Indeed, data on the amount of necrosis could highlight the strength of the colonies' exposure to mechanical impacts and are consequently crucial to study changes in their demographic structure over time. In fact, the size, structures, and the high level of tissue necrosis of the colonies suggest a low conservation status of the studied Tunisian populations.

**Keywords:** Demography; Octocoral; Tunisia; Disturbance levels; *Eunicella singularis*; Scientific diving.

### Introduction

Climate change is a major pressure damaging biological diversity and ecosystems around the world by affecting phenology, physiology, and ecological interactions among species, causing changes in their biogeography and altering community structure and dynamics (Harley *et al.*, 2006; Walther, 2010). Mass mortality events (MMEs) of benthic species have occurred in the last few decades and have been related to marine heat waves linked to global warming (Harley *et al.*, 2006; Halpern *et al.*, 2015; Garrabou *et al.*, 2019; Smale *et al.*, 2019). Overall, more than 90 species of sessile macro-invertebrates were affected in Mediterranean, particularly sponges and gorgonians (Cerrano *et al.*, 2000; Garrabou *et al.*, 2009, 2019). Indeed, gorgonians (Cnidaria, Octocorallia) were the most damaged species, with an average loss of more

than 50% in density and biomass in impacted areas (Linares *et al.*, 2005; Cupido *et al.*, 2008; Coma *et al.*, 2009; Santangelo *et al.*, 2015). Physiological stress induced by climate change-related disturbances, in particular, strong and recurrent marine heat waves (Cerrano *et al.*, 2000; Kersting *et al.*, 2013; Turicchia *et al.*, 2018; Bensoussan *et al.*, 2019) severely threaten these species and increase their vulnerability to pathogens (Rivetti *et al.*, 2014). This vulnerability could be enhanced by decreasing food sources, caused by the stratification of the water column in summer, which seems to be the basis of mass mortality events (MME's) reported for gorgonian species in the last decades in the north-western Mediterranean Sea (Linares *et al.*, 2005; Crisci *et al.*, 2011; Huete-Stauffer *et al.*, 2011; Marbà *et al.*, 2015; Garrabou *et al.*, 2019).

Among affected species, the white gorgonian *Eunicella singularis* (Esper, 1791) suffered different mass

mortalities events the north-western Mediterranean Sea causing important decreases in densities and biomass with rates of up to 90% necrosis (Coma *et al.*, 2006; Garrabou *et al.*, 2009; Gambi *et al.*, 2018; Turicchia *et al.*, 2018). The first event was reported by Harmelin (1984) at La Ciotat in the Ligurian Sea followed by an unprecedented large scale MME in 1999 that impacted thousands of populations in a wide area including the Ligurian Sea, Balearic Islands, and Port Cros National Park (Cerrano *et al.*, 2000; Perez *et al.*, 2000; Coma *et al.*, 2006). Such events have been observed from 2002 to 2017 in many localities but particularly in the Tyrrhenian Sea (Ischia and Procida Islands, Gulf of Naples, Montecristo Island, Tavolara Island), and the northern and south-western Mediterranean Sea (Cabo de Palos, Cabrera Island, Tabarca, Spain and Banyuls sur Mer, France) (Cigliano & Gambi, 2007; Gambi *et al.*, 2006; Garrabou *et al.*, 2009; Carella *et al.*, 2014; Gambi *et al.*, 2018; Turicchia *et al.*, 2018). Being a cold-water species, *E. singularis* is very sensitive to thermal stress (Previati *et al.*, 2010; Linares *et al.*, 2013; Vezzulli *et al.*, 2013) as demonstrated by thermo-tolerance experiments carried out on this species showing that its exposure to temperatures greater than 26°C could have lethal consequences (Coma *et al.*, 2009; Linares *et al.*, 2013) and thus, it can be considered as a potential indicator of climate change impacts on Mediterranean benthic communities (Coma *et al.*, 2006).

*Eunicella singularis* forms dense populations in Mediterranean rocky coastal habitats between a few meters down to 50 m depth, mainly in the western Mediterranean but also in the Adriatic and Aegean seas (Carpine & Grasshoff, 1975; Weinberg, 1976, 1980; Linares *et al.*, 2008; Gori *et al.*, 2011a; Ghanem *et al.*, 2018). *Eunicella singularis* is a photophilic species that inhabits horizontal or subhorizontal substrates and can be also found in coralligenous assemblages. It is also the only Mediterranean gorgonian species hosting symbiotic zooxanthellae (Forcioli *et al.*, 2011).

While our knowledge on population structure, conser-

vation status, life-history traits, and population genetics of long-lived species has improved in the last two decades (Linares *et al.*, 2008; Sini *et al.*, 2015; Ledoux *et al.*, 2018), it remains rare for many key species in the Mediterranean. Such basic information is a crucial prerequisite for the establishment of management measures to ensure the conservation of those organisms. To fill this gap, the main goal of this work was to provide data on demographic characteristics and the disturbance level of *E. singularis*, one of the most common octocoral species in Tunisian waters.

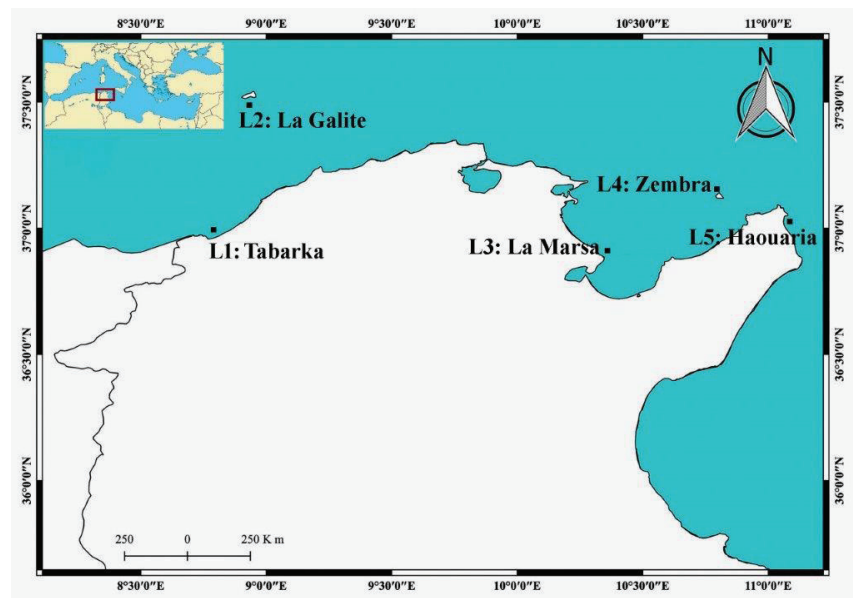
## Materials and Methods

### The species *Eunicella singularis*

*Eunicella singularis* is characterised by a longevity reaching 25-30 years with an average growth of approximately 2-5 cm/year (Weinberg & Weinberg 1979; Munari *et al.*, 2013) displaying higher growth rates during the first year of life (Viladrich *et al.*, 2016). This gorgonian presents a high rate of recruitment with large larval and juvenile mortality (Weinberg & Weinberg, 1979; Linares *et al.*, 2008). According to studies performed in the NW Mediterranean Sea, the species is gonochoric with rare cases of hermaphroditism and reproduces annually in late May and June when the water column is fully stratified (Gori *et al.*, 2007; Ribes *et al.*, 2007). Gametogenesis displays a seasonal pattern of development characterised by a single annual maturation of the gametes (Ribes *et al.*, 2007).

### Sampling design

The white gorgonian displays abundant populations in northern and eastern Tunisian coasts (Ghanem *et al.*, 2018). In order to characterise the population structures



**Fig. 1:** Study sites.

**Table 1.** Characteristics of the sampled populations including coordinates, type of habitat, type of inclination, and protection level).

Locality ID	Locality name	Site	Coordinates	Habitat	Inclination	Protection level	
<b>Sector 1</b>							
L1	Tabarka	Cap Tabarka	37.10694° N	8.81° E	Rocky outcrop	Sub-vertical	Unprotected
		Rocher Mérou	37.14611° N	8.871944° E	Wall	Vertical	Unprotected
L2	La Galite	Pointe Mistral	37.75389° N	8.907222° E	Rocky outcrop	Sub-vertical	MPA
		Les chiens	37.58889° N	8.993056° E	Rocky outcrop	Sub-vertical	MPA
		Gallo	37.70139° N	9.108611° E	Rocky outcrop	Sub-vertical	MPA
		Gallina	37.67556° N	9.770556° E	Wall	Vertical	MPA
<b>Sector 2</b>							
L3	La Marsa	Kobet Hwé	37.46778° N	10.5825° E	Rocky outcrop	Sub-vertical	Unprotected
		Sidi Abdelaziz	37.47944° N	10.605° E	Rocky outcrop	Sub-vertical	Unprotected
L4	Zembra	Capo Grosso	37.21417° N	10.8325° E	Rocky outcrop	Sub-vertical	MPA
		La Cathédrale	37.15528° N	10.87083° E	Small stones	----	MPA
L5	Haouaria	Ras Drak	37.20194° N	11.24722° E	Rocky outcrop	Sub-vertical	Unprotected
		Banc Madyouna 1	37.15639° N	11.27194° E	Rocky outcrop	Sub-vertical	Unprotected
		Banc Madyouna 2	37.15° N	11.27972° E	Rocky outcrop	Sub-vertical	Unprotected

and their conservation status, five localities and 13 sites were studied within two sectors including two Marine Protected Areas (MPA) (Fig. 1 and Table 1). In the north-west sector (1) data from six sites within two localities were collected: Tabarka and La Galite MPA. The north-east sector (2) comprises data from seven sites within three localities: La Marsa, Zembra MPA, and Haouaria.

A scuba diving-based monitoring of the species demography was conducted at depths not exceeding 40 m. Data for each population were collected only once, during the period 2015-2016. In the case of the two MPAs of Zembra and La Galite, a special authorisation was issued by the Ministry of Agriculture permitting field surveys.

### Demographic parameters

In each sector surveyed, the upper distribution limit of the white gorgonian *Eunicella singularis* populations was noted. The assessment of the main population characteristics was carried out following Linares *et al.* (2008). Colony density, height, proportion of injured surface (i.e., tissue necrosis), and type of injury were considered as the main demographic descriptors. Measurements were conducted using 50×50 cm quadrats placed randomly within the *E. singularis* populations. Density was determined based on the number of colonies present within the quadrats. For each colony, maximum height was considered as the distance from the colony base to the tip of the furthest apical branch and measured with a tape measure.

### Assessment of disturbance impact level

Extent of injury per colony, type of injury (see below), and proportion of healthy colonies were monitored to survey the effect of potential disturbances. Percentage of partial and full necrosis for each colony was visually estimated. Extent of injury was defined as the proportion of the denuded colony axis that appeared devoid of coenenchyma tissue and/or that was overgrown by other organisms. Three types of injury were considered on the basis of the presence or absence of different epibionts; Type A: recent injury (up to 1 month) characterised by denuded colony axis; Type B: colony overgrowth by pioneer species (filamentous algae and hydrozoans), signifying injuries of 1-12 months old; Type C: colony overgrowth by algae, bryozoans, and sponges, showing an old injury of approximately > 12 months (Linares *et al.*, 2005). Finally, from the collected information we calculated the percentage of healthy colonies in each population.

A total of 818 colonies was examined. Colonies with < 10% of injured surface were considered as healthy, colonies with injuries ≥ 10% of total surface area were classified as affected, whereas 100% injury corresponded to dead colonies (Garrabou *et al.*, 2009; Linares *et al.*, 2008).

### Data analysis

Height measurements of living colonies (displaying < 100% of injured surface) were grouped into five classes: 1-10, 11-20, 21-30, 31-40, and > 41 cm, and the descriptive distribution parameters of skewness ( $g_1$ ) and kurtosis ( $g_2$ ) were estimated. Skewness ( $g_1$ ) is a measure of the symmetry of a distribution using its mean. If skewness

is significant, the distribution is asymmetrical. Positive skewness describes the dominance of small size classes in the population. Kurtosis ( $g_2$ ) is a measure of the peakiness of a distribution near its central model. A significant kurtosis value indicates that the variable has longer tails than those of a normal distribution and therefore the prevalence of a particular size class in the population. Coefficients of  $g_1$  and  $g_2$  were considered significant if the ratio to their standard error was  $> 2$  (Sokal & Rohlf, 1995). Comparisons were made to determine inter and intra-localities differences using StatGraphics software.

## Results

### Bathymetric distribution range

The upper depth distribution limit of *Eunicella singularis* populations varied among the five localities. For sector 2 (La Marsa locality) some populations appeared at 7 m depth. At Tabarka (L1) and Haouaria (L5) localities, the bathymetric distribution of the species was between 15 and 28 m depth. However, at the two MPAs of Zembra and La Galite, colonies were found from 19 m to 35 m depth.

### Demographic characteristics

A total of 818 colonies were monitored in northern Tunisia (Table 2) including 448 colonies in sector 1 and 370 in sector 2. L1 (Tabarka), L2 (La Galite), and L5 (Haouaria) localities displaying larger abundance comparing to L3 (La Marsa) and L4 (Zembra MPA) where

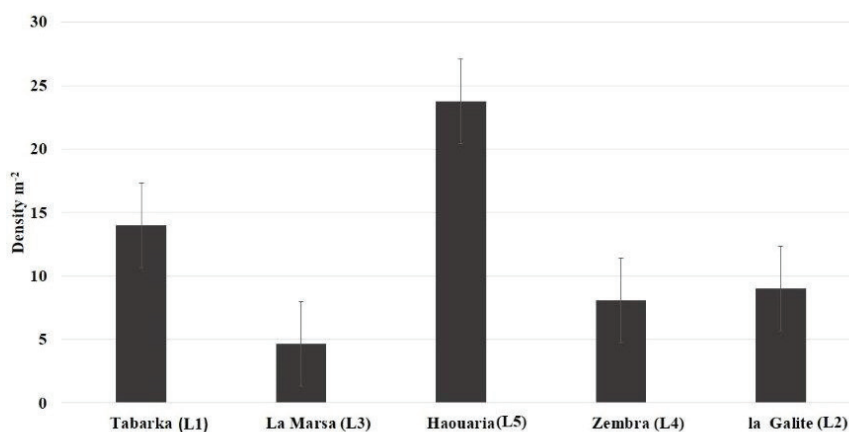
only 27 and 41 colonies were studied, respectively, due to the low abundance of the species at searched depths in these sites.

Mean population density of *E. singularis* was  $11.91 \pm 7.42$  SD (mean  $\pm$  standard deviation) per  $m^2$ . The minimum and the maximum densities were observed in Sector 2 in the locality of La Marsa (L3) with 4.5 colonies per  $m^2$  and Haouaria locality (L5) with 25.86 colonies per  $m^2$ , respectively (Fig.2). Among localities, higher densities were noted in L5 ( $23.77 \pm 2.91$  individuals per  $m^2$ ) followed by L1 (Tabarka:  $14 \pm 11.61$  individuals per  $m^2$ ). However, lowest densities were observed in L2 (La Galite with  $9.03 \pm 3.62$  SD per  $m^2$ ), followed by L4 (Zembra:  $8.09 \pm 2.95$  individuals per  $m^2$ ) and L3 (La Marsa), which represents the most depleted localities in terms of number of colonies present per site ( $4.65 \pm 0.21$  individuals per  $m^2$ ).

ANOVA results suggests that a significant variability in density exists among localities ( $p < 0.05$ ) (Table 2). Among sites, a significant difference in population density exists among both Tabarka (L1) and La Galite (L2) sites ( $p < 0.05$ ).

Overall, mean colony height was  $16.49 \pm 5.59$  cm, while the maximum recorded height was 51 cm (Table 3). Among localities (Fig.3), mean colony height was greater in L5 (Haouaria:  $26.07 \pm 3.29$  cm), followed by L2 (La Galite:  $16.88 \pm 0.96$  cm). The lowest height values were recorded in L1 (Tabarka :  $13.5 \pm 1.01$  cm), L4 (Zembra:  $13.14 \pm 1.42$  cm), and L3 (La Marsa:  $12.85 \pm 1.68$  SD cm). Significant differences in height were observed only at locality level ( $p < 0.05$ ) and at Haouaria sites ( $p < 0.05$ ) (Table 4; ANOVA test).

Since the number of colonies present in some prospected sites is not representative at surveyed depths, height frequency distribution of *E. singularis* populations



**Fig. 2:** Mean density (colonies per  $m^2$ ) of *Eunicella singularis* populations by locality. Whisker span indicates standard deviation.

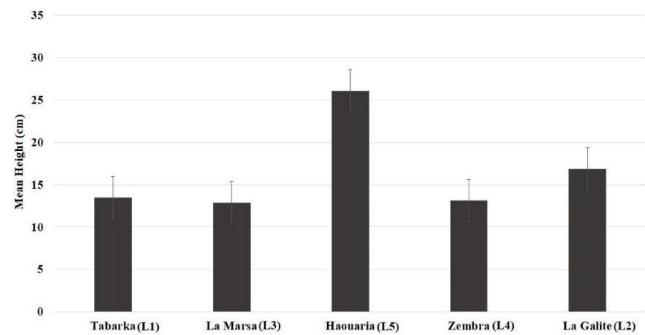
**Table 2.** Summary of ANOVA results for *Eunicella singularis* population density.

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between localities	9762.84	4	2440.71	50.38	0.0000*
Within localities	13953.7	288	48.4505		
<b>Total</b>	<b>23716.6</b>	<b>292</b>			

\*Statistically significant differences ( $p < 0.05$ ).

**Table 3.** Population characteristics of *Eunicella singularis* by locality and site.

Locality ID	Locality name	Site	Depth (m)	Mean density / m <sup>2</sup>	SD	Height (cm)	Number of colonies	Mean height (cm)	SD	Kurtosis	Skewness	
L1	Tabarka	Cap Tabarka	15-18	5.79	2.95	100	5	14.22	0.53	5.32	2.07	
		Rocher Mérou	20-26	22.22	8.80	100	4	12.79	0.55	5.58	1.36	-1.47
L2	Galite	Pointe Mistral	20-24	14	9.09	36	6	18.22	1.33	8	0.81	-0.07
		Les chiens	19-21	8,8	3.77	100	6	16.01	0.56	5.61	1.37	-1.13
		Gallo	24-26	5,33	2	12	6	16.41	3.10	10.77	2.71	3.03
L3	La Marsa	Gallina	20-27	8	4.32	100	7	16.9	0.61	6.16	1.92	-0.12
		Kobet Hwé	7-15	4.5	1.41	9	7	11.66	1.17	3.53	0.78	-0.03
		Sidi Abdelaziz	10-20	4.8	1.65	18	7	14.05	0.96	4.10	0.12	-0.79
L4	Zembra	Capo Grosso	25-30	6	3.88	15	7	12.13	1.00	3.90	0.65	-0.40
		La Cathédrale	30-35	10,18	6.28	26	5	14.15	1.24	6.34	1.54	0.25
L5	Haouaria	Ras Drak	18-24	25	11.4	100	12	23.98	0.72	7.25	1.45	-0.83
		Banc Madyouna (1)	20-26	25.86	10.4	101	12	24.37	0.81	8.24	1.07	-2.13
		Banc Madyouna (2)	20-25	20.44	7.74	101	12	29.87	0.83	8.41	1.48	0.01



**Fig. 3:** Mean colony height of *Eunicella singularis* per locality. Whisker span indicates standard deviation.

was studied only in three localities (L1, L2, and L5). Indeed, the size frequency distribution of populations examined was either asymmetrical or relatively symmetrical depending on the site (Fig. 4). A significant positive skewness, indicating a prevalence of smaller size classes, was found for Tabarka (L1) and Zembra (L4) populations. Among these positively asymmetric populations, the site Cap Tabarka located in L1 has also a significant positive kurtosis value, suggesting a predominance of one or the two smaller size classes (> 0-10 and > 10-20 cm height). Most populations had a non-significant negative kurtosis value. Although a large proportion of large colonies (20-30 cm height) appeared in most sites, the number of colonies larger than 30 cm was generally low.

### Health status

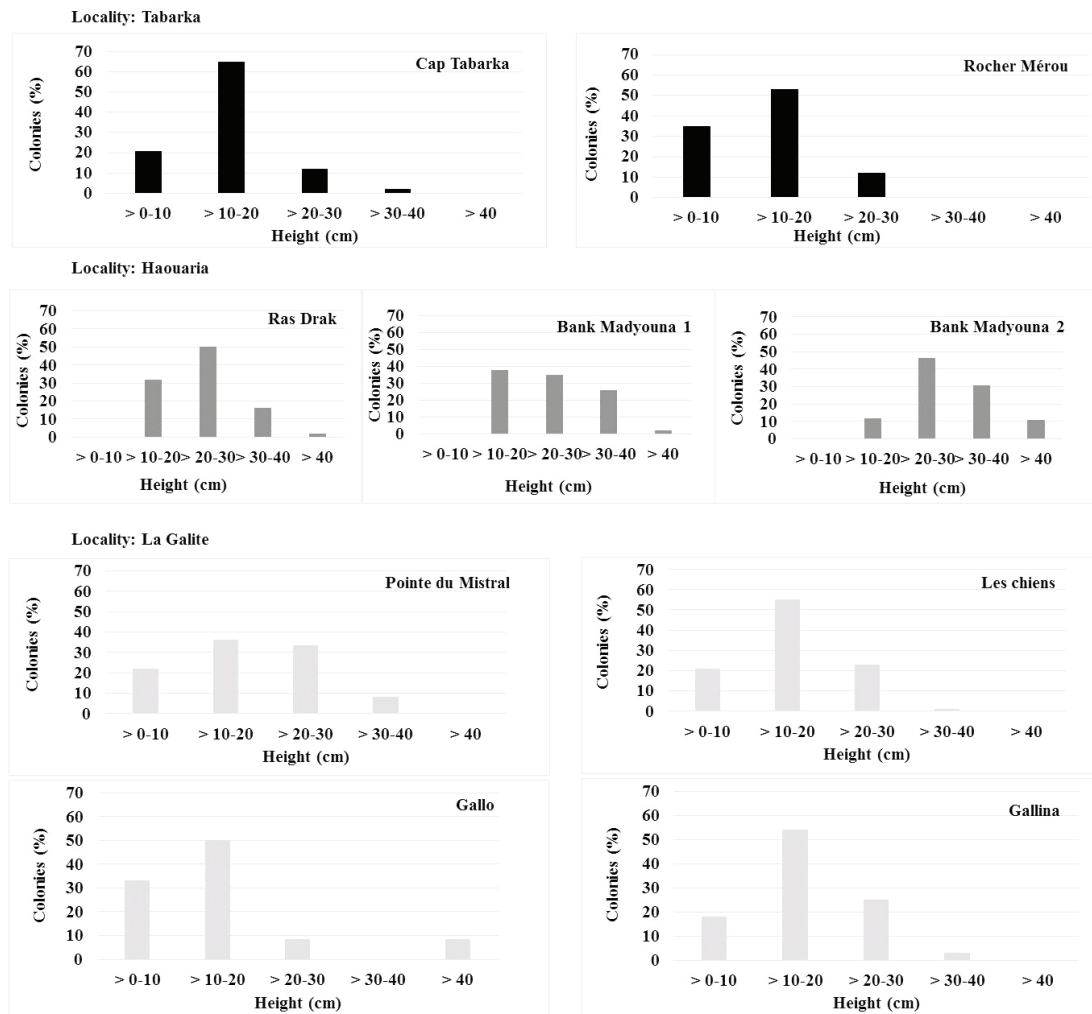
Among populations, the mean extent of injury of gorgonian tissue differs considerably, ranging from 12.47% to 58.88% (Table 5). In Tabarka (L1), 19.2% of the colonies in Cap Tabarka and 24.15% in the Rocher Merou were affected. With regard to the locality of La Marsa (L3), extent of injury varies between 38.66% and 58.88%. At Haouaria (L5), the mean extent of injury was 13.55% ± 1.68. In the MPA of Zembra (L4), the necrosis is estimated as 19.4% ± 4.6 and for the four sites of the La Galite MPA (L2), the mean extent of necrosis is 28.86% ± 17.51.

Six out of thirteen populations have an average extent of injury less than 10% and the remaining populations display extent of injury between 10% and 99% of the total surface: the proportion of affected colonies varies between 28 and 77.77%. The highest values were observed in L3 (La Marsa) (72 and 77%), in Zembra in the Capo Grosso site (60%), and in the MPA of La Galite (L2) with 58% in Gallo, 55% in La Pointe du Mistral, and 53% in Les Chiens site. The percentage of dead colonies is 11.11% in La Marsa (L3), between 2 and 6% in Tabarka (L1), and 1-16.66% in La Galite (L2). Overall, the highest proportions of healthy colonies (< 10% of total injured surface) were recorded in the localities of Haouaria (L5: 60-64%), Zembra (L4) in the site of La Cathédrale (53.85%), and Tabarka (L1) in Cap Tabarka site (56%).

**Table 4.** Summary of ANOVA results for *Eunicella singularis* colony height.

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between Localities	24649.4	4	6162.34	126.83	0.0000*
Within localities	39501.2	813	48.5869		
Total	64150.5	817			

\*Statistically significant differences ( $p < 0.05$ ).



**Fig. 4:** Height frequency distribution of *Eunicella singularis* populations per site, grouped per locality.

Most affected colonies have type B injury (covered by pioneer organisms, therefore a sign of mortality having occurred during the last years) (Fig. 5). In Tabarka (L1), populations showed a proliferation of pioneer species (type B); In Zembra (L4), the site of Capo Grosso had the highest proportion of recent tissue necrosis (type A injury = 20%), and the locality of La Galite (L2) had average levels of all types of lesions, except in Gallo and Gallina sites, where recent injuries are very low. Since the sampling time was not concomitant between the localities, statistical comparison is not applicable in this specific case.

## Discussion

This study provides the first assessment of the population structure and conservation status of the white gorgonian *Eunicella singularis* in the south Mediterranean Sea, particularly in Tunisia. This work focused only on shallow populations despite the presence of the species down to 60 m depth (Ghanem *et al.*, 2018). The spatial distribution of *E. singularis* depends on the combined effects of biological and environmental factors (Weinberg, 1980; Zabala & Ballesteros, 1989). Indeed, it was usually observed on rocky outcrops or on steep slopes (Linares *et al.*, 2008; Gori *et al.*, 2011a, b). Results are in accordance with other studies on the bathymetric distribution of these populations, which have very variable upper and lower

**Table 5.** Summary data on injury characteristics of *Eunicella singularis* colonies per locality and site.

Locality ID	Locality Name	Site	Depth (m)	N	Extent of colony injury (%)		Proportion of uninjured, injured and dead colonies			Proportion of Colonies per type of injury		
					Mean	SD	< 10%	≥ 10% - ≤ 99%	100 %	A	B	C
L1	Tabarka	Cap Tabarka	15-18	100	19.2	29.5	56	42	2	15	23	6
		Rocher Mérou	20-26	100	24.1	32.6	45	49	6	17	23	15
L2	Galite	Pointe Mistral	20-24	36	21.6	28	44.4	55.5	0	11.1	22.2	22.2
		Les chiens	19-21	100	28	32	46	53	1	11	23	20
		Gallo	24-26	12	53.3	38.6	25	58.3	16.6	0	41.6	33.3
L3	La Marsa	Gallina	20-27	100	12.4	22.9	72	28	0	2	20	6
		Kobet Hwé	7-15	9	58.8	31.4	11.1	77.7	11.1	0	66.6	22.2
L4	Zembra	Sidi Abdelaziz	10-20	18	31.6	28.5	27.7	72.2	0	16.6	38.8	16.6
		Capo Grosso	25-30	15	22.6	23.4	40	60	0	20	6.6	33.3
L5	Haouaria	La Cathédrale	30-35	26	16.1	19.4	53.8	46.1	0	7.6	38.4	0
		Ras Drak	18-24	100	12	19.3	63	37	0	11	20	6
		Banc Madyouna (1)	20-26	101	12.4	19.5	64.3	35.6	0	7.9	9.9	6.9
		Banc Madyouna (2)	20-25	101	15.4	23.1	60.4	39.6	0	15.8	14.8	8.9

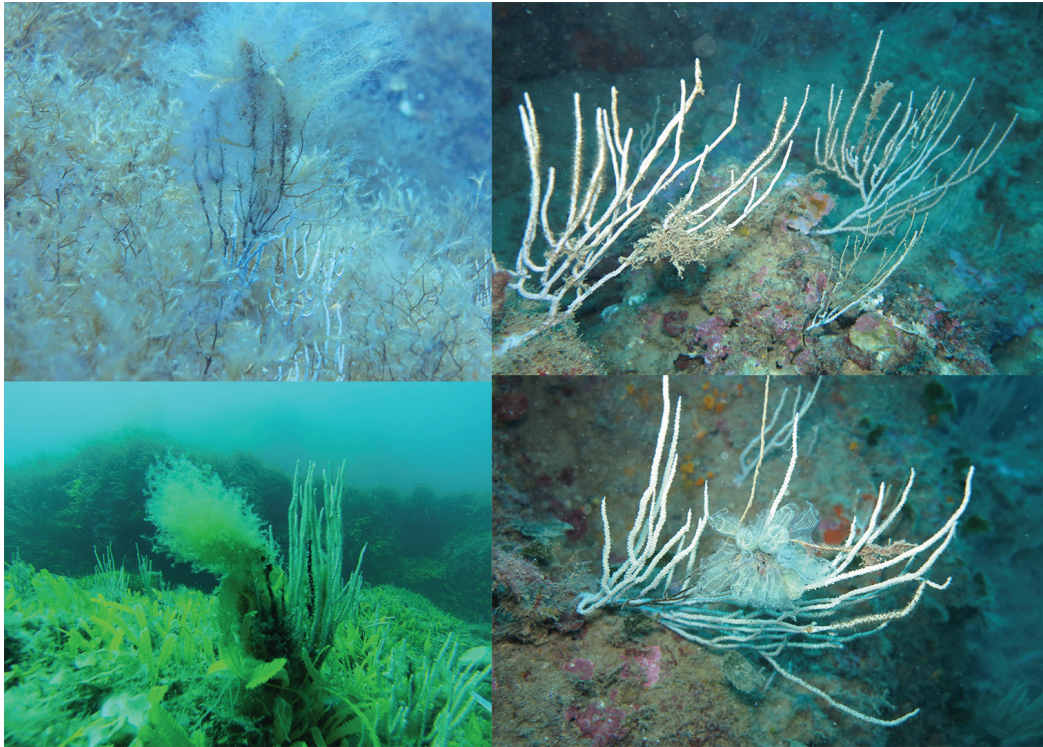
depth limits (Linares *et al.*, 2008; Gori *et al.*, 2011b). In fact, Tunisian populations were found at depths ranging from 7 m (this study) to 60 m depth (Ghanem *et al.*, 2018) and this large fluctuation suggests the tolerance of the species to a wide spectrum of environmental conditions, especially a wide range of light intensity, which may be between 3% and 44% of surface light (Weinberg, 1976; Linares *et al.*, 2008).

Density values of the Tunisian populations are very low compared to those observed in north-western Mediterranean (Linares *et al.*, 2008; Hereu *et al.*, 2014). Mean population density of *E. singularis* was approximately 12 colonies per m<sup>2</sup> in Tunisia with extremes ranging from 1 to 48 colonies per m<sup>2</sup> whereas the densities observed in the N-W Mediterranean are of the order of 20 per m<sup>2</sup> and ranging from 1 to 56 gorgonians / m<sup>2</sup>. In contrast, the height of the colonies of white gorgonians is approximately 15 cm in the N-W Mediterranean and slightly higher (16.5 cm high) for the Tunisian colonies.

Injuries in gorgonians are either generated by mechanical abrasion or predation, which are usually low in natural conditions, or through necrosis of tissue under stressed conditions, such as thermal anomalies, causing partial or total mortality of colonies. Affected colonies may shatter, regenerate, or become colonised by epibionts (Coma *et al.*, 2006). *Eunicella singularis* has been affected by several MMEs related to temperature anomalies in the N-W Mediterranean, some of which have led to high levels of tissue necrosis and significant population declines (Garrabou *et al.*, 2009; Gambi *et al.*, 2018; Turicchia *et al.*, 2018). Although injury estimates were made only once at each site in this study, they do, however, provide baseline data regarding the conservation status for future monitoring. Our assessment emphasised that the extent of injury of colony tissue is relatively high for most studied sites, except for the locality of Haouaria with the percentage of affected colonies being lower than 50% in the northern and eastern sectors of Zembra and Tabarka (Table 2). The large proportion of affected colonies suggests that the majority of *E. singularis* populations observed were impacted by severe disturbances. This study highlights that the Tunisian populations have more necrosis than those observed in other sites in the N-W Mediterranean (Linares *et al.*, 2008; Hereu Fina *et al.*, 2014) but were similar to the necrosis values exhibited by populations affected by mass mortality events (Cerrano *et al.*, 2000; Linares *et al.*, 2005; Coma *et al.*, 2006). Nevertheless, according to Ben Mustapha & El Abed (2000), the populations of *E. singularis* in the locality of Tabarka (L1) showed 70% necrosis while colonies displaying total mortality numbered no less than 30%, while in the present study the same populations had a total mortality rate of below 6% and less than 50% of colonies were affected by necrosis. These results suggest that during the last decades the populations at this locality have improved their conservation status.

Most populations have colonies with a high degree of injury (between 10% and 99%) and especially old necrosis (types B and C). Epibionts develop on the bare axes of colonies affected by mortality, so that their presence is an indicator of past disturbance (Coma *et al.*, 2004; Linares





**Fig. 5:** Affected colonies of the white gorgonian *Eunicella singularis* in different studied sites.

*et al.*, 2005; Coma *et al.*, 2006). The population of Gallo (L2) has the highest proportion of dead colonies and also wounds with a naked axis, indicating high levels of recent mortality, possibly induced by local environmental factors such as the change in currents (Andromède, 2010). Most populations show both recent and old necrosis, suggesting that the impact of recent and past mass mortality events such as those of 1999 and 2003 in the southern Mediterranean is more pronounced and has significantly affected gorgonian species. In addition, the proportions of colonies with bare axes and apical tips were higher than that observed in populations of the Mediterranean coasts of Spain and France (Linares *et al.*, 2008; Hereu Fina *et al.*, 2014) highlighting the impact of recent stressors.

Several direct and indirect anthropogenic stressors act upon Mediterranean gorgonian populations, namely destructive fishing practices, anchoring, unregulated recreational diving, mucilaginous algal aggregates, invasive algae, and mass mortality outbreaks caused by abnormal seawater temperature increases, and most of these interact with additive and synergistic effects (Harmelin & Marinopoulos 1994; Mistri & Ceccherelli, 1996; Bavestrello *et al.*, 1997; Garrabou *et al.*, 2009; Cebrian *et al.*, 2012; Linares *et al.*, 2013).

Basic quantitative information necessary for assessing the conservation status of *Eunicella singularis* populations in Tunisian waters is absent. This work highlights that the rates of necrosis are relatively homogenous among sites. Indeed, and according to the available temperature data for the MPA of Zembra ([www.t-mednet.org](http://www.t-mednet.org)), the position of the seasonal thermocline reaches 25 m and temperatures can exceed 28° C. As a result, it is possible that the deep shift of the seasonal thermocline could be a significant factor affecting *E. singularis* populations.

Populations of the white gorgonian evaluated in this study were dominated by medium size classes ranging between 10 and 20 cm (first reproductive colonies), and only one locality (Haouria L5) had a population size prevalence of between 20 cm and 30 cm height. Our results do not match with previous work carried out in the Mediterranean, where a dominance of small colonies was observed (Coma *et al.*, 2006; Linares *et al.*, 2008; Gori *et al.*, 2011a). These previous studies suggest that the high abundance of small colonies was explained by a high sustained recruitment over time and high survival rate of *E. singularis* juveniles (Ribes *et al.*, 2007; Linares *et al.*, 2008). In contrast, the dominance of average sizes for most Tunisian populations could be explained by a recruitment limitation and probable low survival of juveniles related to mass mortalities episodes, with significant necrosis decreasing the reproduction effort in the species (Coma *et al.*, 2006). In addition, also suggested by Coma *et al.* (2006), small colonies can suffer more from partial mortality than large colonies, which could be a reason for the absence of small-sized colonies in Tunisian gorgonians. Another stressor causing this recruitment limitation could be the presence of invasive algae, which have been demonstrated to negatively affect the recruitment and juvenile survival of this species (Linares *et al.*, 2012).

This study of the demography and the conservation status of the populations of *E. singularis* in Tunisia could be a basis for future work on this species showing a high artificiality in comparison to other studied populations from the NW Mediterranean Sea. However, it is important to highlight the case of Haouria with the highest population density, greatest mean height, high proportions of large colonies, and the lowest levels of tissue damage. Their populations may deserve some conservation measures given that this location is an unprotected

area, in order to prevent potential stressors that may affect their well-conserved populations at least in the context of Tunisian waters. Further knowledge of the biology and ecology of this gorgonian in all its spatial range is needed to characterise future trends in a climate change scenario. Indeed, the monitoring of these populations could contribute towards improving our knowledge regarding the dynamics of the species over longer time scales and to understand impacts of global changes and therefore propose conservation plans and management policies.

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